ARCHIVES

OF

INTERNAL MEDICINE

EDITORIAL BOARD

N. C. GILBERT, Chief Editor, Chicago

ARTHUR BLOOMFIELD, San Francisco

REGINALD FITZ, Boston

J. H. MUSSER, New Orleans

RUSSELL M. WILDER, Rochester, Minn.

VOLUME 64

1939

PUBLISHERS

AMERICAN MEDICAL ASSOCIATION

CHICAGO, ILL.

Medical
Wahrs

CONTENTS OF VOLUME 64

JULY 1939. NUMBER 1

PAGE

Haverhill Fever: Report of a Case with Review of the Literature. Elliston Farrell, M.D.; George H. Lordi, M.D., and Joseph Vogel, Brooklyn... 1

A Review of Meningitis Due to Micrococcus Tetragenus: Report of One Case with Bacteriologic Study. Thorburn S. McGowan, M.D., Manila, Philippine Islands, and Paul Kisner, M.D., Fort Leavenworth, Kan..... 15

Experimental Production of Neutropenia with Aminopyrine. E. M. Butt, M.D.; A. M. Hoffman, M.D., and S. N. Soll, M.D., Los Angeles..... 26

Anatomic Observations on Seventy Hospital Patients After Sudden Death. James R. Lisa, M.D., and James Finlay Hart, M.D., New York..... 43

Roentgenograms of the Chest and the Intracutaneous Tuberculin Test for Adults: A Comparative Study. Luke W. Hunt, M.D., Chicago..... 49

Test of Glomerular Function with Sodium Ferrocyanide: Further Studies. Edward J. Stieglitz, M.D., Garrett Park, Md..... 57

Renal Function and the Number of Glomeruli in the Human Kidney. J. M. Hayman Jr., M.D.; J. W. Martin Jr., M.D., and Max Miller, M.D., Cleveland 69

Bacterial Endocarditis: Report of a Case in Which the Cause Was Actinomyces Bovis. Nathaniel Uhr, M.D., New York..... 84

Protamine Zinc Insulin: A Metabolic Study; Treatment in Two Cases of Severe Diabetes by Equally and Unequally Divided Diets, with Comments on Criteria for Treatment. Edward Tolstoi, M.D., and Frederick C. Weber Jr., M.D., with the Technical Assistance of Vincent Toscani, B.S., New York..... 91

Total, Differential and Absolute Leukocyte Counts and Sedimentation Rates Determined for Healthy Persons Nineteen Years of Age and Over. Edwin E. Osgood, M.D.; Inez E. Brownlee, B.A.; Mable W. Osgood, B.A.; Dorothy M. Ellis, B.A., and William Cohen, M.D., Portland, Ore. 105

Limitations of Biopsy of Sternal Marrow. E. V. Kandel, M.D., and G. V. LeRoy, M.D., Chicago..... 121

Metastatic Calcification: Report of Two Cases. David M. Grayzel, M.D., Ph.D., and Max Lederer, M.D., Brooklyn..... 136

Progress in Internal Medicine:

Blood: A Review of the Literature (Concluded). Cyrus C. Sturgis, M.D.; Raphael Isaacs, M.D.; S. Milton Goldhamer, M.D., and Frank H. Bethell, M.D., Ann Arbor, Mich..... 148

News and Comment..... 213

Book Reviews..... 214

AUGUST 1939. NUMBER 2

Effect of Dihydrotachysterol in Treatment of Parathyroid Deficiency. Edward Rose, M.D., and F. William Sunderman, M.D., Ph.D., Philadelphia.... 217

Evaluation of Sulfanilamide in the Treatment of Patients with Subacute Bacterial Endocarditis. Wesley W. Spink, M.D., and F. Hughes Crago, M.D., Minneapolis 228

Myocardial Infarction Without Significant Lesions of Coronary Arteries. Harry Gross, M.D., and William H. Sternberg, M.D., New York..... 249

Hemolytic Jaundice: Immediate and Delayed Changes in the Blood After Splenectomy. John C. Sharpe, M.D.; Charles W. McLaughlin Jr., M.D., and Ralph Cunningham, M.D., Omaha..... 268

Polyostotic Fibrous Dysplasia: Report of a Case. Thomas Horwitz, M.D., and Abraham Cantarow, M.D., Philadelphia..... 280

Boeck's Sarcoid: Autopsy in a Case with Visceral Lesions. E. F. Cotter, M.D., Nashville, Tenn..... 286

AUGUST—*Continued*

PAGE

Cardiac Sequelae of Embolism of the Pulmonary Artery. Henry Horn, M.D.; Simon Dack, M.D., and Charles K. Friedberg, M.D., New York 296

Calcium and Digitalis Synergism: The Toxicity of Calcium Salts Injected Intravenously into Digitalized Animals. Paul K. Smith, Ph.D.; Alexander W. Winkler, M.D., and Hebbel E. Hoff, Ph.D., M.D., New Haven, Conn. 322

Relation of Myasthenia Gravis to Hyperthyroidism. Melvin W. Thorner, M.D., Philadelphia 330

Heart Failure in Subacute Bacterial Endocarditis. William C. Buchbinder, M.D., and Otto Saphir, M.D., Chicago 336

Clinical Significance of Variations in Serum Phosphatase in Hepatic and Biliary Disorders. Louis Winkelman, M.D., and Albert Schiffmann, M.D., Brooklyn 348

Progress in Internal Medicine:
Infectious Diseases. Hobart A. Reimann, M.D., Philadelphia 362

Book Reviews 406

News and Comment 408

SEPTEMBER 1939. NUMBER 3

Range of Normal Blood Pressure: A Statistical and Clinical Study of 11,383 Persons. Samuel C. Robinson, M.D., and Marshall Brucer, Chicago 409

Bilirubin and Urobilin Content of Bile Obtained by Duodenal Drainage: Normal Values and Values for Patients with Cholecystitis. M. Royer, M.D., Buenos Aires, Argentina 445

"Cardiac Cirrhosis" of the Liver: A Clinical and Pathologic Study. Herbert M. Katzin, M.D.; John V. Waller, M.D., and Herrman L. Blumgart, M.D., Boston 457

Hematology of Sternal Marrow and Venous Blood of Pregnant and of Nonpregnant Women. Harry H. Pitts, M.D., and Evelyn A. Packham, R.N., Vancouver, British Columbia, Canada 471

Phosphatase Activity in Chronic Arthritis. Charles Leroy Steinberg, M.D., and Louise Catherine Suter, Rochester, N. Y. 483

Electrocardiographic Findings in Cases of Ventricular Aneurysm. Maurice Eliaser Jr., M.D., and Jerome Konigsberg, M.D., San Francisco 493

Effects on the Cardiovascular System of Man of Fluids Administered Intravenously: III. Studies of the Glomerular Filtration Rate as Measured by the Urea Clearance. D. Rourke Gilligan, M.S.; Mark D. Altschule, M.D., and Arthur J. Linenthal, A.B., Boston 505

Experimental Renal Insufficiency Produced by Partial Nephrectomy. Alfred Chanutin, Ph.D., and Stephan Ludewig, Ph.D., University, Va.:
XI. Diets Containing Dried Extracted Liver 513
XII. Diets Containing Dried Extracted Meat 526

Intracerebral Carcinomatous Metastases. Clarence C. Hare, M.D., and Gabriel A. Schwarz, M.D., New York 542

Primary Carcinoma of the Liver: Tumor Thrombosis of the Inferior Vena Cava and Right Auricle. Raymond Gregory, M.D., Washington, D. C. 566

Thiocyanate Dermatitis: Report of a Case. Mervin E. Green, M.D., and James S. Snow, M.D., Ann Arbor, Mich 579

Lipoid Pneumonia: Report of Two Cases. Curtis F. Garvin, M.D., Cleveland 586

Progress in Internal Medicine:
Vascular Diseases: A Review of Some of the Recent Literature, with a Critical Review of the Surgical Treatment. George W. Scupham, M.D.; Géza de Takáts, M.D.; Theodore R. Van Dellen, M.D., and William C. Beck, M.D., Chicago 590

News and Comment 656

Book Reviews 657

OCTOBER 1939. NUMBER 4

PAGE

Reflex Coronary Artery Spasm Following Sudden Occlusion of Other Coronary Branches. G. W. Manning, M.A.; C. G. McEachern, M.D., B.Sc. (Med.), and G. E. Hall, M.D., Ph.D., Toronto, Canada 661

Angina Pectoris in Hereditary Xanthomatosis. Carl Müller, M.D., Ullevaal, Norway 675

Chronic Leukemia: The Early Phase of Chronic Leukemia, the Results of Treatment and the Effects of Complicating Infections; a Study of Eighty-Six Adults. Maxwell M. Wintrobe, M.D., and L. Lee Hasenbush, M.D., Baltimore 701

Traumatic Rupture of the Pericardium: Study of Twenty-Two Cases, with Two and One-Half Year Period of Survival in One Case; Review of Literature. S. F. Crynes, M.D., and Warren C. Hunter, M.D., Portland, Ore. 719

Experimental Renal Insufficiency Produced by Partial Nephrectomy. Alfred Chanutin, Ph.D., and Stephan Ludewig, Ph.D., University, Va.:

XIII. A Summary of the Effect of Whole Liver, Whole Meat, Extracted Liver and Extracted Meat Diets on Renal Hypertrophy, Renal Function, Blood Pressure and Cardiac Hypertrophy 747

XIV. Diets Containing Whole Dried Yeast 756

Age, Sex and Hypertension in Myocardial Infarction Due to Coronary Occlusion. A. M. Master, M.D.; Simon Dack, M.D., and H. L. Jaffe, M.D., New York 767

Juvenile Diabetes Mellitus. William H. Grishaw, M.D.; Howard F. West, M.D., and Bertnard Smith, M.D., Los Angeles 787

Hyperthermia, Genuine and Spurious. Ward J. MacNeal, M.D., New York 800

Prolonged Hyperthermia: Report of a Case with Necropsy. Ward J. MacNeal, M.D.; Henry H. Ritter, M.D., and S. Milton Rabson, M.D., New York 809

Multiple Myeloma Associated with Nodular Deposits of Amyloid in the Muscles and Joints and with Bence Jones Proteinuria. Leonard Tarr, M.D., and Henry W. Ferris, M.D., New York 820

Progress in Internal Medicine:

Gastroenterology: A Review of the Literature from July 1938 to July 1939. Chester M. Jones, M.D., Boston 834

Book Reviews 888

News and Comment 891

Obituaries:

William J. Mayo, M.D 892

Charles H. Mayo, M.D 895

NOVEMBER 1939. NUMBER 5

Optimum Time for Administration of Protamine Zinc Insulin. Morton F. Mark, M.D., New York 897

Deleterious Effects of Experimental Protamine Insulin Shock. James W. Sherrill, M.D., and Eaton M. MacKay, M.D., San Diego, Calif 907

Transport of Air Along Sheaths of Pulmonic Blood Vessels from Alveoli to Mediastinum: Clinical Implications. Charles C. Macklin, M.D., Ph.D., F.R.S.C., London, Ontario, Canada 913

Studies on "Essential" Hypertension: I. Classification. Henry A. Schroeder, M.D., and J. Murray Steele, M.D., New York 927

Shock. L. Meyler, M.D., Groningen, Netherlands 952

Incidence of Fatal Cardiovascular Disease in Charleston, S. C., with Particular Reference to Hypertension. Thomas M. Peery, M.D., Washington, D. C., and S. M. Langsam, M.D., New York 971

Blood "Guanidine": Further Observations. Ralph H. Major, M.D.; C. J. Weber, M.D., Ph.D., and M. J. Rumold, M.D., Kansas City, Kan 988

Multiple Myeloma. Helmuth Ulrich, M.D., Boston 994

NOVEMBER—*Continued*

PAGE

Lipocaic and Fatty Infiltration of the Liver in Pancreatic Diabetes. Lester R. Dragstedt, M.D.; Cornelius Vermeulen, M.D.; W. Carter Goodpasture, M.D.; Paul B. Donovan, Ph.D., and William A. Geer, M.D., Chicago 1017

Structural Changes in the Lungs of Drug Addicts. George C. Cole, M.D., New York 1039

Progress in Internal Medicine:
Syphilis: A Review of the Recent Literature. Joseph Earle Moore, M.D., and Charles F. Mohr, M.D., Baltimore 1053

News and Comment 1128

Notices:
Cumulated Index of the Archives 1128

Book Reviews 1129

DECEMBER 1939. NUMBER 6

Periarteritis Nodosa: Report of a Case. Reginald Fitz, M.D.; Harry Parks, M.D., and Charles F. Branch, M.D., Boston 1133

Chronic Malaria: A Clinical Consideration. George H. Fondé, M.D., and Edgar C. Fondé, M.D., Mobile, Ala 1156

Biophotometer Test as Index of Nutritional Status for Vitamin A. Grace Steininger, M.S., and Lydia J. Roberts, Ph.D., Chicago 1170

Infantilism in Ulcerative Colitis. Sidney Davidson, M.D., New York 1187

Recurring Encephalomeningoradiculitis with Fibromyositis Following Poliomyelitis: A Bacteriologic Study of Sixty-Four Cases. Edward C. Rosenow, M.D., Rochester, Minn 1197

Tumors of the Adrenal Glands: I. A Modified Air Injection Roentgen Technic for Demonstrating Cortical and Medullary Tumors. Oliver Cope, M.D., and Richard Schatzki, M.D., Boston 1222

Elevation of Urea Nitrogen Content of the Blood Following Hematemesis or Melena. Leon Schiff, M.D., and Richard J. Stevens, M.D., with the Technical Assistance of Ellen S. Garber, M.S., Cincinnati 1239

Intercapillary Glomerulosclerosis: A Syndrome of Diabetes, Hypertension and Albuminuria. Robert A. Newburger, M.D., and John P. Peters, M.D., New Haven, Conn 1252

Physiologic Basis of Intravenous Dextrose Therapy for Diseases of the Liver. Samuel Soskin, M.D., and Mayer Hyman, M.D., Chicago 1265

Studies on Destruction of Red Blood Cells: I. Chronic Hemolytic Anemia with Paroxysmal Nocturnal Hemoglobinuria; an Investigation of the Mechanism of Hemolysis, with Observations on Five Cases. Thomas Hale Ham, M.D., Boston 1271

Diagnosis of Hypersensitiveness to the Bee and to the Mosquito with Report on Successful Specific Treatment. Robert L. Benson, M.D., Portland, Ore. 1306

Progress in Internal Medicine:
Review of Neuropsychiatry for 1939. Stanley Cobb, M.D., Boston 1328

Book Reviews 1340

News and Comment 1342

Notices:
Cumulated Index of the Archives 1342

General Index 1343

ARCHIVES of INTERNAL MEDICINE

VOLUME 64 JULY 1939 NUMBER 1

COPYRIGHT, 1939, BY THE AMERICAN MEDICAL ASSOCIATION

HAVERHILL FEVER

REPORT OF A CASE WITH REVIEW OF THE LITERATURE

ELLISTON FARRELL, M.D.

GEORGE H. LORDI, M.D.

AND

JOSEPH VOGEL

BROOKLYN

Haverhill fever is characterized by "an abrupt onset . . . a rubelliform to morbilliform eruption . . . chiefly on the extremities . . . and an inflammation of the joints with marked pain and tenderness."[1] The diagnosis is made by isolation, from blood or from joint fluid, of a highly pleomorphic filamentous or spindle-shaped aerobic or microaerophilic gram-negative organism, Haverhillia multiformis,[2] which requires serum for growth in artificial mediums. Recently, in the medical service of the Long Island College Hospital, a patient was observed who presented the clinical picture of Haverhill fever. Haverhillia multiformis was isolated from the blood stream on seven occasions.

REPORT OF CASE

A. S., a 40 year old Italian woman, was admitted to the medical service of the Long Island College Hospital on Nov. 19, 1937, complaining of pain and swelling of the joints of about five days' duration.

She stated that three weeks before admission she had been bitten on the web between the fourth and the fifth finger of the left hand by a young rat encountered in the kitchen garbage receptacle. The wound bled at the time. The patient washed the hand and applied iodine to the bleeding area. Complete healing followed, and the history of this trauma was not elicited until nearly a month after admission.

About one week before the patient's entry a slight cough and a sore throat developed. A sharp chill occurred, lasting about fifteen minutes. That evening the patient felt feverish and had pain in both knees and both ankles. For the

From the Department of Medicine and the Department of Bacteriology, the Long Island College of Medicine.

1. Place, E. H., and Sutton, L. E.: Erythema Arthriticum Epidemicum (Haverhill Fever), Arch. Int. Med. **54**:659 (Nov.) 1934.

2. Parker, F., Jr., and Hudson, N. P.: The Etiology of Haverhill Fever (Erythema Arthriticum Epidemicum), Am. J. Path. **2**:357, 1926.

next four days she was confined to bed with headache, repeated vomiting and pain in the neck and back. A rash appeared around the knees and ankles.

Two days before admission the woman's condition improved sufficiently for her to get up, but a return of symptoms soon forced her again to bed. There now developed pain and swelling in the right shoulder, elbow and wrist. There was an increase in the pain and swelling of the knees and ankles.

Ten years before admission the patient had had pains of the joints with slight fever but was not confined to bed. Her general health otherwise had been good.

On physical examination the temperature was 103 F., the pulse rate 128, the respiratory rate 28 and the blood pressure 110 systolic and 70 diastolic. The woman seemed acutely ill. The throat was injected. The left lobe of the thyroid was slightly enlarged. The heart showed no murmurs, but there was some roughening of the first sound at the apex, and the second pulmonary sound was accentuated at the base. The abdomen was moderately tender in the right upper quadrant. The spleen was not felt. The neck showed pain on flexion. The right elbow and wrist were swollen, tender and painful when moved. The left wrist was tender but not swollen. Both knees were swollen and painful on movement, the right more than the left. Fluid was present in the right knee. The muscles of the back were tender. A maculopapular eruption was present on the extensor aspects of the lower parts of the legs. The diagnosis on admission was acute rheumatic fever.

The hemoglobin content of the blood was 70 per cent (Sahli); red cells numbered 4,190,000 and white cells 11,450, with 87 per cent polymorphonuclears. The sedimentation time was thirty-seven minutes (18 mm.). The urine contained no albumin or casts. The blood contained 130 mg. of sugar and 8 mg of urea nitrogen per hundred cubic centimeters. The Wassermann and Kahn reactions of the blood were negative.

A roentgenogram of the chest taken on November 22 showed no evidence of active tuberculosis, but there were increase in interstitial fibrosis and evidence of fibrous pleuritis on the right.

Course in the Hospital.—As shown in the chart (fig. 1), the patient's temperature during the first week in the hospital (second week of the disease) fluctuated between 99.4 and 104.6 F. before assuming a continuous level of about 100 F. Except for irregular elevation above that point during the third and fourth weeks in the hospital (fourth and fifth weeks of the disease), there was a gradual decline to a normal level over a total febrile period of about eight weeks.

The arthritis present on the patient's admission slowly subsided until December 22, when there was a return of pain in the right knee and left shoulder. This exacerbation lasted about four weeks; efforts at relief of pain were largely ineffectual, as in the first attack. Pain on motion, tenderness and swelling characterized the arthritic manifestations. Fluid was thought to be present only once: in the right knee joint on the patient's admission. The fluid was not aspirated. At various times roentgen study of the right elbow, both shoulders, the lumbar portion of the spine, the sacroiliac joints and the hip joints gave negative results.

The eruption seemed to be of embolic rather than allergic type. It was maculopapular and was confined to the extremities. New lesions appeared from time to time during the illness, and some of these were frankly petechial, being red and tender at first, with a central pinpoint of hemorrhage. Several such lesions appeared on the fingers. The cutaneous lesions never suppurated but subsided slowly, leaving pigmented areas which later underwent desquamation.

During this illness the patient's general condition remained surprisingly good. Apart from the pain and discomfort of the arthritis, she suffered little incon-

venience from the fever or other accompaniments of the infection. It was found that with exercise the typical signs of mitral stenosis could be elicited. It was felt that the woman had an old rheumatic lesion referable to her earlier attacks of arthritis.

Treatment included the administration of salicylates, which was largely ineffective, and of codeine, which gave fairly good results. During a period of six days, from December 11 through December 16, a total of 12.7 Gm. of sulfanilamide was given without apparent influence on the course of the infection. Because of its reported efficacy in actinomycosis,[3] administration of 1 Gm. of thymol daily was started on Jan. 17, 1938, and continued with brief intermissions for about three

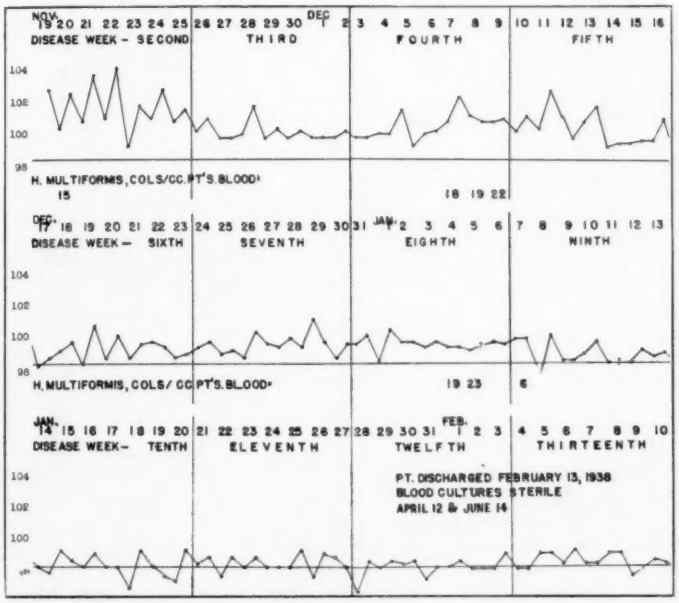


Fig. 1.—Chart of temperature (F.) in a case of Haverhill fever. Only readings taken at 8 a. m. and 8 p. m. are shown.

weeks. After introduction of this drug the fever remained below 100 F., but the medication was begun late in the course of the disease, and the clinical impression was that it probably had no specific action.

Course After Leaving the Hospital.—The patient was discharged from the hospital on February 13, having been completely afebrile for about two weeks. There was still some disability in the left shoulder, but the other joints were apparently normal in appearance and in function. After a period of relative freedom from symptoms, pain recurred in the left shoulder and to a lesser extent in

3. Myers, H. B.: Thymol Therapy in Actinomycosis, J. A. M. A. **108**:1875 (May 29) 1937.

the left elbow. This was noted during May. During the latter part of June diathermy was used with apparent success. At the time of writing there has been no recurrence of the acute arthritic symptoms, other than that just noted, nor has there been a return of the cutaneous lesions which were present on the patient's admission to the hospital.

Bacteriology.—Haverhillia multiformis was recovered from the blood in pure culture on seven occasions while the patient was in the hospital: on Nov. 20, 1937, 15 colonies per cubic centimeter; on December 7, 18 colonies; on December 8, 19 colonies; on December 9, 22 colonies; on Jan. 4, 1938, 19 colonies; on January 5, 23 colonies, and on January 7, 6 colonies. Subsequent to the patient's discharge cultures of blood taken on April 12 and June 14 were sterile.

The micro-organisms isolated corresponded in every particular tested with the organism of Parker and Hudson.[2] The marked pleomorphism was its most striking characteristic (fig. 2). Coccobacilli, rods, filaments and threads were all on occasions encountered in a single oil immersion field, and round or fusiform swellings situated in any portion of the rod were frequently seen. Distinct branching was noted. The micro-organism varied in length from 2 to 15 microns and in width from 0.2 to 0.5 micron. The diameter of the swellings ranged from 2 to 6 microns. In blood broth the rods and filaments were often seen in tangled masses.

Haverhillia multiformis is gram-negative, not acid-fast and not encapsulated. No motility was observed in hanging drop or in dark field preparations.

On whole human blood agar, as well as in 0.1 per cent dextrose beef heart infusion broth (p_H 7.8) to which human blood had been added, the organism grew extremely well. The optimal temperature range for growth was 35 to 38 C. Good growth was obtained aerobically. Only fair proliferation was obtained on chocolate agar, and no growth occurred on plain agar or in 0.1 per cent dextrose beef infusion broth. Blood, serum, ascitic fluid or egg yolk furnished the indispensable growth factors.

In serum or ascitic fluid broth the growth appeared at the bottom of the tube as a gray-white layer made up of small particles of aggregated organisms. Colonies developing in the broth on the side of the tube appeared as soft white fluffy aggregates which were not easily broken up. The supernatant broth remained clear, and no pellicle was observed.

The surface colonies on semisolid mediums were round, regular, soft and moist, ranging from 1 to 2 mm. in diameter. Usually they were discrete, but on very moist mediums some confluence was observed.

Intraperitoneal inoculation of 2 white mice with 0.5 cc. of a forty-eight hour blood broth culture resulted in the death of one mouse in twenty-four hours and of the other mouse in forty-eight hours. After death Haverhillia multiformis was seen in direct smear and recovered in pure culture from the peritoneal fluid and from the heart's blood.

The patient's serum collected in the fifth week of disease agglutinated her own organism completely in dilutions ranging from 1:20 to 1:640.

References to other cultural characteristics are available in the article of Parker and Hudson[2] and the book of Topley and Wilson.[4] One interesting characteristic of the organism is its specific pathogenicity for

4. Topley, W. W. C., and Wilson, G. S.: The Principles of Bacteriology and Immunity, Baltimore, William Wood & Company, 1936, pp. 274 and 997.

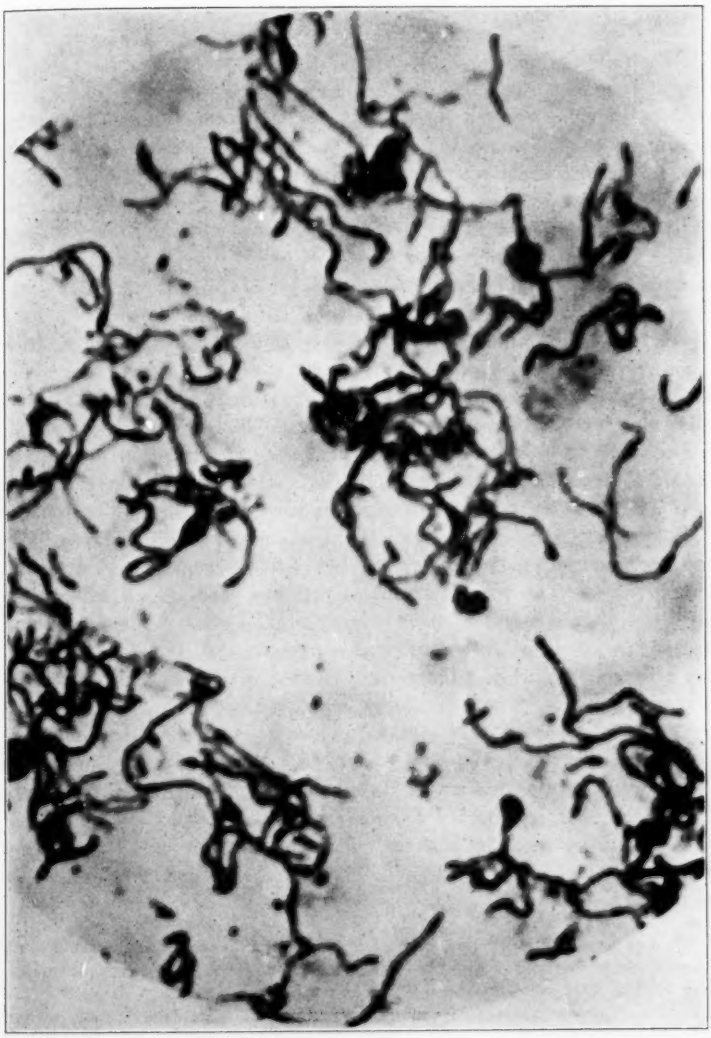


Fig. 2.—Haverhillia multiformis (Parker and Hudson[2]) obtained by forty-eight hour culture in whole blood agar. Magnification, × 2,700. Note streptobacillary and branching forms.

mice, in which it causes an epizootic, with arthritis as a prominent manifestation of the infection.[5]

HISTORICAL REVIEW

The condition we have just described was apparently first recognized by Schottmüller[6] in 1914. Since this time other cases have been reported which were not linked with his, and other names have been assigned both to the clinical syndrome and to the organism. As a result, case reports are now hidden in the literature under a variety of headings. It is instructive to bring together these scattered case reports for comparison with our own, as presented in the accompanying table. In our series we include only those cases in which an organism closely resembling Haverhillia multiformis (Parker and Hudson, 1926[2]) has been demonstrated on culture of blood or joint fluid.

In 1914 Schottmüller[6] reported 2 cases of *Bisskrankheit,* but positive blood cultures were obtained only in the first of these cases; hence only the first case will be considered here. In the patient, a laboratory *Diener,* there developed, following rat bite, a local lymphadenitis and lymphangitis with fever and considerable pain and tenderness in muscles of the left arm and leg. Movement of the left shoulder caused exquisite pain. It is interesting that the bite occurred not on the left but on the right thumb. A roseola-like exanthem was observed on the arms, legs and abdomen seven days after the bite. This eruption faded within sixteen days. The patient's temperature was 39.3 C. (102.7 F.) on admission to the hospital, seven days after the bite. It was remittent, tending to fall to a lower level each successive day until the sixteenth day after the bite, when a sharp rise occurred, accompanied by exacerbation of symptoms. The temperature subsequently fell slowly to a normal level, and the patient was eventually discharged well after a stay of nearly ten weeks in the hospital. Eight blood cultures taken during this period were positive for an organism Schottmüller named Streptothrix muris ratti. Growth occurred, apparently under aerobic conditions, on Löffler's serum medium and on milk agar but not on other culture mediums. The organism was described as gram-positive (*nach der Gramschen Methode . . . blau tingiert*).

---

5. (*a*) Levaditi, C.; Selbie, R. F., and Schoen, R.: Le rhumatisme infectieux spontané de la souris provoqué par le Streptobacillus moniliformis, Ann. Inst. Pasteur **48**:308, 1932. (*b*) Mackie, T. J.; van Rooyen, C. E., and Gilroy, E.: An Epizootic Disease Occurring in a Breeding Stock of Mice: Bacteriological and Experimental Observations, Brit. J. Exper. Path. **14**:132, 1933. (*c*) van Rooyen, C. E.: The Biology, Pathogenesis and Classification of Streptobacillus Moniliformis, J. Path. & Bact. **43**:455, 1936. (*d*) Topley and Wilson.[4]

6. Schottmüller, H.: Zur Aetiologie und Klinik der Bisskrankheit, Dermat. Wchnschr. (supp.) **58**:77, 1914.

Sporadic Instances of Haverhill Fever Proved by Cultivation of an Organism Resembling Haverhillia Multiformis from Peripheral Blood or from Joint Fluid

Report				Patient			Disease			Characteristics of Organism			
Case No.	Year	Author	Locality	Sex	Age	Occupation	Apparent Mode of Infection	Designation Given Disease	Name Given Organism	Pleomorphism	Gram Stain	Growth on Ordinary Mediums	Outcome
1	1914	Schottmüller.....	Hamburg, Germany	Male	25 yr.	Laboratory servant	Rat bite	Bisskrankheit	Streptothrix muris ratti	+	+	—	Recovered
2	1916	Blake.....	Boston	Female	67 yr.	Shopkeeper	Rat bite	Rat bite fever	Streptothrix muris ratti	+	—	—	Died; autopsy
3	1917	Litterer.....	Nashville	Male	14 yr.?	.....	Rat bite	Rat bite fever	Streptothrix longus	+	±	—	Recovered
4	1917	Litterer.....	Nashville	Male	5 yr.	.....	Rat bite	Rat bite fever	Streptothrix brevis	+	±	?	Recovered
5	1918	Dick and Tunnicliff..	Chicago	Male	10 yr.	.....	Weasel	Typical of rat bite fever	Streptothrix putorii	+	—	+	Recovered
6	1918	Tunnicliff and Mayer	Chicago	Female	13 days	.....	Rat bite	Rat bite fever	Closely related to Streptothrix putorii	+	—	±	Died; autopsy
7	1925	Ebert and Hesse.....	Leningrad, U.S.S.R.	Female	42 yr.	Teacher	Rat bite	Japanisches Rattenbissfieber	Streptothrix muris ratti	+	?	—	Recovered
8	1925	Levaditi, Nicolau and Poincloux	.....	.....	.....	.....	Obscure	Érythème polymorphe aigu	Streptobacillus moniliformis	+	—	±	Recovered
9	1926	Dodd.....	Baltimore	Female	10 yr.	.....	Rat bite	Erythema arthriticum epidemicum	Not identified but from description streptothrix	+	—	—	Recovered
10	1929	Teissier, Rivalier, Reilly and Garnier	Paris, France	Female	35 yr.	Writer	Obscure	Érythème infectieux	Streptobacillus moniliformis	+	—	±	Recovered
11	1932	Hazard and Goodkind	Boston	Male	58 yr.	.....	Obscure	Haverhill fever	Haverhillia multiformis	+	—	—	Recovered
12	1934	Scharles and Seastone	Boston	Male	25 yr.?	Medical student	Rat bite	Haverhill fever	Haverhillia multiformis	+	—	—	Recovered
13	1937	Lemierre, Reilly, LaPorte and Morin	Paris, France	Male	40 yr.	Railway worker	Rat bite	Fièvre par morsure de rat	Streptobacillus moniliformis	+	—	±	Recovered
14	1938	Farrell, Lordi and Vogel	Brooklyn	Female	40 yr.	Housewife	Rat bite	Haverhill fever	Haverhillia multiformis	+	—	—	Recovered

In 1916 Blake [7] reported a fatal infection with Streptothrix muris ratti following rat bite. Autopsy disclosed acute ulcerative endocarditis of the mitral valve, with infarcts lodged in the spleen and kidney. The organism was recovered from the blood of the patient during life and from heart's blood post mortem. In the same year Tileston [8] reported 2 cases of "rat-bite fever," in each case basing his diagnosis on the history of rat bite and on the clinical course, which was characterized by periodic fever. In 1 case he observed rodlike forms in the peripheral blood during the febrile paroxysms, but failure to cultivate this possible "streptothrix" necessitates exclusion of the report from this series.

In 1917 Litterer [9] reported 2 additional cases, in both of which the disease followed rat bite and in both of which he established the diagnosis by blood culture. Minor differences in the organisms recovered led him to assign different specific names to the two strains he isolated, but their essential similarity with Schottmüller's organism is apparent from his descriptions.

In 1918 Dick and Tunnicliff [10] and Tunnicliff and Mayer [11] reported the fifth and sixth cases of apparent Haverhillia multiformis infection. In 1 of their cases the disease followed weasel bite; in the other, in which death ensued and autopsy was done, it occurred after rat bite. From blood culture, in both instances a "streptothrix" was grown, variations in the cultural characteristics of which again led to new terminology, which is listed in the table.

In 1925 Ebert and Hesse [12] reported isolation of an organism showing great similarity (*grosse Ähnlichkeit*) to the organisms of Schottmüller and of Blake in a case of a disease they described as *japanisches Rattenbissfieber* (sodoku). In the same year Thorp [13] gave an extremely incomplete account of a case of fever following rat bite, with isolation of a "leptothrix" in material from swellings which developed on each wrist and near the sternoclavicular joints. The case is not included in our series because there is no report of blood culture and the history is inadequate.

---

7. Blake, F. G.: The Etiology of Rat-Bite Fever, J. Exper. Med. **23**:39, 1916.

8. Tileston, W.: The Etiology and Treatment of Rat-Bite Fever, J. A. M. A. **66**:995 (April 1) 1916.

9. Litterer, W. A.: Study of the Streptothrix Isolated in Two Cases of Rat-Bite Fever, Tr. Sect. Path. & Physiol., A. M. A., 1917, p. 275.

10. Dick, G. F., and Tunnicliff, R.: A Streptothrix Isolated from the Blood of a Patient Bitten by a Weasel (Streptothrix Putorii), J. Infect. Dis. **23**:183, 1918.

11. Tunnicliff, R., and Mayer, K. M.: A Case of Rat-Bite Fever, J. Infect. Dis. **23**:555, 1918.

12. Ebert, B., and Hesse, E.: Zur Klinik und Bakteriologie des japanischen Rattenbissfiebers (Sodoku), Arch. f. klin. Chir. **136**:69, 1925.

13. Thorp, E.: Rat-Bite Fever in an Infant, Brit. M. J. **2**:255, 1925.

In 1925 [14] (final report, 1926 [15]) Levaditi, Nicolau and Poincloux described a case in which the clinical course was characterized by remittent fever with a four to six day cycle. There was no history of rat bite. Three febrile paroxysms were experienced, following each of which there appeared a maculopapular eruption, chiefly on the limbs. Severe pain developed in the left ankle and in the right acromioclavicular joint. Recovery was complete. On two occasions there was isolated in blood culture an organism they named Streptobacillus moniliformis. No reference was made to Schottmüller's organism or to the series of cases which had been appearing as instances of rat bite fever. Attention was focused on the dermatologic aspects of the infection, and the condition was described as acute erythema multiforme (*érythème polymorphe aigu*).

These 8 case reports described sporadic instances of Haverhillia multiformis infection, but in 1926 Place, Sutton and Willner [16] under the designation erythema arthriticum epidemicum (final report by Place and Sutton, 1934 [1]) reported an epidemic of 86 cases of what seems to have been the same infection, and in 12 of these cases Parker and Hudson isolated from blood or from joint fluid the organism they named Haverhillia multiformis: "slender, gram-negative and non acid resisting rods staining with some difficulty; often forming threads and showing tendency toward branching; marked irregularity of form with swellings and enlargements; fermentation of some carbohydrates; in general, requiring blood or ascitic fluid for growth." [2] These epidemic cases were not associated with rat bite and were not at the time identified with the earlier isolated cases we have cited. Dodd,[17] however, in the same year (1926), under the name erythema arthriticum epidemicum, reported a case of disease following rat bite which she had observed in Baltimore in 1923, making a retrospective diagnosis. The organism was incompletely studied.

In 1929 Teissier, Rivalier, Reilly and Garnier [18] reported a case of "infectious erythema" with isolation of Streptobacillus moniliformis on blood culture. There was no history of rat bite.

---

14. Levaditi, C.; Nicolau, S., and Poincloux, P.: Sur le rôle étiologique de streptobacillus moniliformis (nov. spec.) dans l'érythème polymorphe aigu septicémique, Compt. rend. Acad. d. sc. **180**:1188, 1925.

15. Levaditi, C.; Nicolau, S., and Poincloux, P.: Recherches sur l'étiologie de l'érythème polymorphe aigu; son agent étiologique, Streptobacillus moniliformis, Presse méd. **34**:340, 1926.

16. Place, E. H.; Sutton, L. E., and Willner, O.: Erythema Arthriticum Epidemicum: Preliminary Report, Boston M. & S. J. **194**:285, 1926.

17. Dodd, K.: An Isolated Case of Erythema Arthriticum Epidemicum, Boston M. & S. J. **194**:633, 1926.

18. Teissier, P.; Rivalier, E.; Reilly, J., and Garnier, G.: Sur un cas d'réythème infectieux du au "Streptobacillus moniliformis," Congrès de dermatologistes et syphiligraphes de langue franç., Paris, Masson & Cie, 1929, p. 73.

In 1932 Hazard and Goodkind [19] gave the first complete report of a sporadic case of a disease identified at the time as Haverhill fever (erythema arthriticum epidemicum) with recovery of Haverhillia multiformis on blood culture. They obtained no history of rat bite, but in 1934 Scharles and Seastone [20] described a case of rat bite infection in a medical student in which diagnosis had been made by cultivation of Haverhillia multiformis from joint fluid.

The most recent case report previous to our own came from France, that of Lemierre, Reilly, LaPorte and Morin,[21] who in 1937 described a case of *fièvre par morsure de rat* with recovery from the blood stream of Streptobacillus moniliformis. Infection with Levaditi's organism was thus finally allied with the *Bisskrankheit* of Schottmüller.

COMMENT

Bacteriology.—Our first question is, are all these organisms the same? Without type cultures for comparison or cross immunity tests, no proof, of course, can be offered; nor will proof of this kind ever be available in all the cases of this series. A number of authors [22] have regarded Streptothrix muris ratti (Schottmüller, 1914), Streptobacillus moniliformis (Levaditi and others, 1926) and Haverhillia multiformis (Parker and Hudson, 1926) as closely related if not identical strains. Others [23] have identified Streptobacillus moniliformis with Haverhillia multiformis without reference to Schottmüller's organism. The fact that his organism was gram-positive and the other organisms in this series gram-negative seems sufficiently explained by Litterer,[9] who remarked that the variability in staining reaction seemed dependent on the age of the culture. Slight morphologic distinctions or differences in sugar fermentation reactions seem unimportant when contrasted with the two major characteristics common to this group: extreme pleomorphism of an unusual type and special growth requirements. All these organisms, except that of Dick and Tunnicliff,[10] can fairly be

19. Hazard, J. B., and Goodkind, R.: Haverhill Fever (Erythema Arthriticum Epidemicum): A Case Report and Bacteriologic Study, J. A. M. A. **99**:534 (Aug. 13) 1932.

20. Scharles, F. H., and Seastone, C. V., Jr.: Haverhill Fever Following Rat-Bite, New England J. Med. **211**:711, 1934.

21. Lemierre, A.; Reilly, J.; Laporte, A., and Morin, M.: Sur une nouvelle fièvre par morsure de rat, Bull. Acad. de méd., Paris **117**:705, 1937.

22. (*a*) Dienes, L., and Edsall, G.: Observations on the L-Organism of Klieneberger, Proc. Soc. Exper. Biol. & Med. **36**:740, 1937. (*b*) Place and Sutton.[1] (*c*) Topley and Wilson.[4]

23. Levaditi, C.: À propos de l'étiologie de l'érythème polymorphe infectieux, contagieux et épidemique, Presse méd. **36**:65, 1928. van Rooyen.[5c] Teissier and others.[18] Lemierre and others.[21]

described in the words of Parker and Hudson [2] as "in general, requiring blood or ascitic fluid for growth."

Three conflicting views have been taken regarding classification of this unusual organism. Schottmüller regarded it as an actinomyces, as did Parker and Hudson, and Topley and Wilson unhesitatingly assigned it to this order. Levaditi placed it among the bacteria, while Klieneberger,[24] in work recently supported by Dienes and Edsall,[22a] maintained that the form previously regarded as single in truth consists of a streptobacillus growing in symbiosis with a rounded, coccoid, "pleuro-pneumonia-like organism" [24a] the "L_1 organism of Klieneberger." [22a] The latter symbiont forms the knobs and fusiform swellings hitherto looked on simply as pleomorphic changes. Van Rooyen,[6c] like Parker and Hudson,[2] stressed the organism's special growth requirements but failed to find true branching. He stated that he looked on it as a single pleomorphic organism of the family Bacteriaceae, tribe Haemophileae, genus Haverhillia, type species Haverhillia multiformis (Parker and Hudson). The literature as we interpret it gives priority to Schottmüller's name for this organism, but as his term, Streptothrix muris ratti, is now unacceptable for taxonomic reasons, the term of Parker and Hudson seems more satisfactory until the difficult question of classification has been authoritatively decided.

Clinical Syndrome.—The reports summarized here have fallen into three clinical groups, and it seems convenient to discuss them under the designations originally applied.

Rat Bite Fever: Of the cases reviewed, in 10 there was a history of rat bite and in 1 of weasel bite; of these cases, 7 were looked on as instances of rat bite fever, Schottmüller carefully naming the condition in his case simply "bite disease." Continued use of the term "rat bite fever" to describe instances of Haverhillia multiformis infection is ill advised, for the term is also in use to describe Spirillum minus infection,[25] or sodoku. The etiologic agents of these two diseases are distinct: the one, Haverhillia multiformis, being a nonmotile pleomorphic bacillus which grows on serum mediums, the other, a highly motile spirillum of uniform morphologic character which has never [26] been

24. Klieneberger, E.: (*a*) The Natural Occurrence of Pleuro-Pneumonia-Like Organisms in Apparent Symbiosis with Streptobacillus Moniliformis and Other Bacteria, J. Path. & Bact. **40**:93, 1935; (*b*) Further Studies on Streptobacillus Moniliformis and Its Symbiont, ibid. **42**:587, 1936.

25. Robertson, A.: Spirillum Minus (Carter 1887), the Aetiological Agent of Rat-Bite Fever: A Review, Ann. Trop. Med. **24**:367, 1930. McDermott, E. M.: Rat-Bite Fever: A Study of the Experimental Disease, with a Critical Review of the Literature, Quart. J. Med. **21**:433, 1928.

26. Bayne-Jones, S.: Rat-Bite Fever in the United States, Internat. Clin. **3**: 235, 1931.

successfully cultivated outside the animal body. Either infection may follow rat bite, but while Haverhillia multiformis infection has occurred without a history of rat bite, no example of Spirillum minus infection has been found in which a history of antecedent rat bite or an account of close contact with rats or with associated animals was lacking. Haverhill fever has caused at least one epidemic; sodoku has never occurred except in sporadic form. Haverhill fever is accompanied by painful arthritis and is resistant to treatment; sodoku is not characterized by articular involvement, and arsenical therapy has a specific curative effect. Both infections have cutaneous manifestations,[27] but one is essentially embolic, the other primarily allergic. Haverhill fever may be characterized by a continued remittent fever or by an intermittent fever with a four to six day periodicity. The febrile reaction in sodoku is characteristically only of the latter type. No one has explained why the temperature charts of these two infections with entirely different causal agents should at times exactly resemble each other, particularly when the fever is of a distinctive, periodic type found rarely, if at all, in any other clinical condition. The type of fever is so unusual that in the past numerous authors have considered a history of rat bite followed by an intermittent fever with a four to six day periodicity sufficient grounds for a case report labeled "Rat Bite Fever." Coincident infection with Haverhillia multiformis and Spirillum minus is theoretically possible,[12] but it has never been proved. Mackie and McDermott [28] suggested the possibility in their case, but examination of their data shows that they were dealing with some organism other than Haverhillia multiformis in association with the spirillum of sodoku. Elucidation of the exact bacteriologic relation between Haverhillia multiformis and Spirillum minus and of the clinical affinities of Haverhill fever and sodoku must await longer observation. Sufficient knowledge is at hand, however, to differentiate the two bacteriologically, and it is unfortunate that three American medical textbooks published during 1937 and 1938 confuse the bacteriologic entity of Spirillum minus infection with infection due to Haverhillia multiformis because of the common association with rat bite and certain clinical similarities.

Erythema (Polymorphous, Infectious, Arthritic): Levaditi and his co-workers [14] were the first to consider Haverhillia multiformis infection as one of the erythemas; but Place, Sutton and Willner [16] independently adopted the same point of view, and the cases of the

27. O'Leary, P. A.: The Dermatologic Aspects of Rat-Bite Fever, Arch. Dermat. & Syph. **9**:293 (March) 1924.

28. Mackie, T. J., and McDermott, E. N.: Bacteriological and Experimental Observations on a Case of Rat-Bite Fever: Spirillum Minus, J. Path. & Bact. **29**: 493, 1926.

Haverhill epidemic were originally described by them as examples of erythema arthriticum epidemicum. An important American textbook of medicine published in 1937 classifies Haverhillia multiformis infection with the erythemas, listing it with types such as erythema nodosum and erythema infectiosum. This conception of the disease as a dermatologic condition is inadequate; the cutaneous eruption is simply one manifestation of primary septicemia, and the disease is not just a local condition but a general systemic infection.

Haverhill Fever: This term was in popular use to describe the cases of the Haverhill epidemic before the cause of the disease was known. We have preferred it because it properly emphasizes the systemic nature of the infection and because it is applicable in sporadic as well as epidemic cases.

Epidemiology.—There are two known modes of infection with Haverhillia multiformis: rat bite [6] and ingestion of raw milk [1] which has been contaminated with the organism. Rats are known to harbor the organism in the nasopharynx [29] as a benign inhabitant or in the lungs [30] as an apparent pathogen associated with bronchopneumonia. An obvious possibility in accounting for the Haverhill epidemic is that rats had access to the milk supply.

Differential Diagnosis.—The history of rat bite or the periodic fever may suggest sodoku, diagnosis of which requires demonstration of Spirillum minus in the patient's blood or in the material aspirated from the local lesion at the site of the rat bite or from the regional lymph nodes. Inoculation of 1 cc. of whole blood intraperitoneally or subcutaneously into white mice free [31] from Spirillum minus infection will ordinarily, after an incubation period of ten days or longer, result in a blood stream infection that will continue for months, and diagnosis can readily be made by dark field examination. In Haverhillia multiformis infection the fever may be of the continued type and may resemble the fever of any infectious or septic condition. The arthritis may suggest undulant fever or rheumatic fever or may be regarded as infectious arthritis of another type. The eruption may be confused with one of the erythemas (infectious erythema or erythema multi-

29. Strangeways, W. I.: Rats as Carriers of Streptobacillus Moniliformis, J. Path. & Bact. **37**:45, 1933. Lemierre and others.[21]

30. Tunnicliff, R.: Streptothrix in Bronchopneumonia of Rats Similar to That in Rat-Bite Fever, J. Infect. Dis. **19**:767, 1916. Klieneberger, E., and Steabben, D. B.: On a Pleuropneumonia-Like Organism in Lung Lesions of Rats, J. Hyg. **37**:143, 1937.

31. Knowles, R.; Das Gupta, B. M., and Sen, S.: Natural Spirillum Minus Infection in White Mice, Indian M. Gaz. **71**:210, 1936. Francis, E.: Rat-Bite Fever Spirochetes in Naturally Infected White Mice, Mus Musculus, Pub. Health Rep. **51**:976, 1936.

forme), or the entire picture may be attributed to one of the eruptive fevers. The final diagnosis rests on culture of blood or of joint fluid.

Treatment.—Neoarsphenamine has been used with apparent success in cases of Haverhillia multiformis infection with periodic fever. In 2 cases in which there was periodic fever the condition responded to autogenous vaccine, according to Litterer.[9] In cases in which there was continued fever, no therapy seems to have been effectual. If, as some have maintained, the organism is one of the actinomyces, further trial of thymol in larger doses seems justifiable.

SUMMARY

Clinical and bacteriologic findings in the fourteenth sporadic case of Haverhill fever to be reported are presented.

Previous case reports are reviewed.

The bacteriology, clinical syndrome, epidemiology, differential diagnosis and treatment of this disease are discussed.

Dr. M. L. Rakieten first suggested the identity of the organism isolated in the case we have reported.

The photograph is the work of Mr. S. Montes.

34 Prospect Park, West.

142 Joralemon Street.

A REVIEW OF MENINGITIS DUE TO MICROCOCCUS TETRAGENUS

REPORT OF ONE CASE WITH BACTERIOLOGIC STUDY

THORBURN S. McGOWAN, M.D.
Passed Assistant Surgeon, United States Public Health Service
MANILA, PHILIPPINE ISLANDS
AND
PAUL KISNER, M.D.
First Lieutenant, Medical Corps Reserve, United States Army
FORT LEAVENWORTH, KAN.

Although Gaffke described Micrococcus tetragenus in phthisical sputum in 1881, the finding of this organism in the spinal fluid is still rare. A search of all the indexed journals discloses only 7 prior cases in the accumulated world literature. Probably the diagnosis of meningitis due to this organism is frequently missed in this country because the symptoms, physical and laboratory findings and clinical course are similar to those of meningococcic meningitis. Reimann [1] in 1934 stated that there were only 2 or 3 reports of any form of infection by M. tetragenus in American literature, in contrast to 170 cases reported in European publications. In recording this case, the eighth, and analyzing the preceding cases, we hope to emphasize sufficiently the similarity to infection with meningococci or staphylococci and to point out the inherent characteristics peculiar to M. tetragenus. A realization of these facts should result in the recognition of the condition in an increasing number of cases in the future.

From the United States Penitentiary Hospital, through the courtesy of Dr. O. C. Williams, United States Public Health Service, Medical Officer in Charge. The bacteriologic studies were carried out in the laboratory of the Station Hospital, Fort Leavenworth, Kan., Captain D. C. Kuhns, Medical Corps, United States Army, in charge.

1. Reimann, H. A.: (*a*) Micrococcus Tetragenus Infection: Review of Literature and Report of a Case, J. Clin. Investigation **14**:311 (May) 1935; (*b*) Bacterial Type Dissociation of Micrococcus Tetragenus Infection: II. Description of Variant Forms, J. Bact. **31**:385 (April) 1936; III. Immunologic Studies of Variant Forms, and Discussion, ibid. **31**:407 (April) 1936; Bacterial Type Transformation: Micrococcus Tetragenus Infection, ibid. **33**:499 (May) 1937; Variation of Micrococcus Tetragenus, Proc. Soc. Exper. Biol. & Med. **34**:344 (April) 1936; Transformation of Bacterial Types, ibid. **35**:64 (Oct.) 1936.

It is well known that M. tetragenus is a common inhabitant of the nose, throat and sputum of patients with tuberculosis. Although it is frequently seen in smears of material from the throat or of sputum, it has been regarded as a nonpathogenic organism; nevertheless, it becomes invasive when the general resistance is lowered. In our case meningitis is thought to have followed invasion of the blood stream by the organism, despite a negative blood culture and the possibility of direct extension from the nasopharynx.

REPORT OF CASE

Clinical History.—On July 16, 1936, D. H., an inmate of the United States Penitentiary, a white man aged 20, lapsed into unconsciousness suddenly while working in the "yard gang." Since the weather had been extremely humid for a number of days, with daily temperature peaks over 100 F., it was assumed on his admission to the prison hospital that he was suffering from heat exhaustion. He soon revived on symptomatic treatment and stated that he had had pains in the neck and general malaise for ten days. Aside from several minor injuries, the past history was irrelevant.

Physical Examination.—At the time of admission he appeared comatose and acutely ill. Neurologic examination showed hyperactive cremasteric, abdominal and Brudzinski reflexes on the right side, contrasted with hypoactive reflexes on the left. The muscles of the neck were rigid, and a definite Kernig sign was present. The pupils were equal and regular and reacted to light. All other physical findings were normal. The nasal and pharyngeal mucous membranes were not inflamed. The clinical impression was heat exhaustion.

Course of the Disease.—On the morning following the admission stiffness of the neck, headache and abnormal reflexes had disappeared; the patient was conscious and rational and, but for a slight elevation of temperature (38 C.) [100.4 F.], appeared to be recovering from his supposed attack of heat exhaustion.

In the afternoon of the second day in the hospital the picture changed. The temperature rose to 38.7 C. (101.7 F.), and the patient began to complain of a bitemporal headache, with pains in the back of his neck. A Kernig and a Brudzinski sign reappeared at the same time. The symptoms now were indicative of definite meningeal involvement, and since there had recently been a case of meningococcic meningitis in the hospital it was assumed that the meningococcus was again the causative organism.

Treatment and Laboratory Findings.—A lumbar puncture was done; the spinal fluid was cloudy and under increased pressure (280 mm. of water). This observation enhanced the assumption that the condition was of meningococcic origin; accordingly, 30,000 units of antimeningococcus serum was given intrathecally in divided doses, with 20 cc. of antitoxin intramuscularly, during the next twelve hours. For the next seven days 10,000 units of the antiserum and 10 cc. of antitoxin were given daily. Spinal fluid was drained eleven times thereafter at appropriate intervals for the relief of pressure symptoms.

Examination of the original specimen of spinal fluid showed a cell count of 5,300 per cubic millimeter, with a differential count of 94 per cent neutrophils and 6 per cent lymphocytes; the reaction for globulin to Pandy's reagent was 4 plus; quantitative determinations showed a decrease in sugar and the colloidal gold test resulted in a curve typical of purulent meningitis (0000244444). A direct smear with Gram stain revealed diplococci, some of which were gram-positive and resembled meningococci in structure but were not intracellular. Cultures made on this and the following two days gave pure growths of gram-positive tetrads.

Serum Reaction.—There is no specific antiserum for M. tetragenus; since the organism was agglutinated with dilutions up to 1:180 by polyvalent antimeningococcus serum, administration of antiserum was continued up to the ninth day,

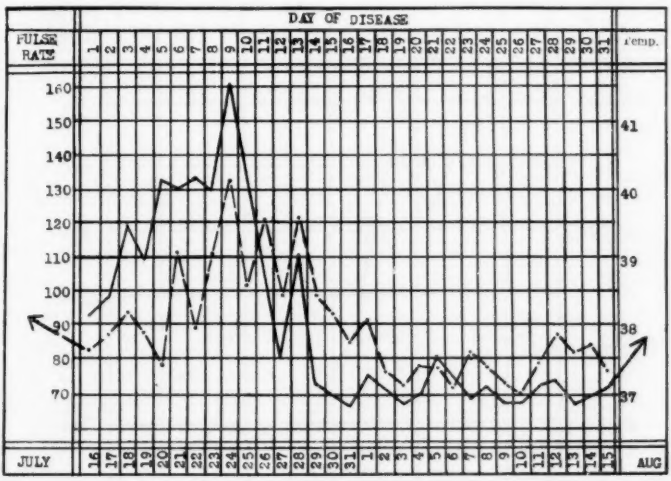


Chart 1.—Pulse rate and temperature.

when suddenly difficulty in breathing and a rapid, thready pulse developed. In the course of the day there appeared petechiae on the lower extremities, general adenitis, an urticarial rash over the entire body, functional heart murmur and cyanosis. The patient became irrational and semicomatose. These findings were interpreted as manifestations of serum sickness. Although the prognosis appeared grave, he improved rapidly; his temperature receded to normal on the fifteenth day and remained within normal limits thereafter until the day of discharge, August 28. Since then he has been normal in every way.

It will be noted in chart 1 that the temperature corresponded to that in cases of typical meningococcic meningitis. It varied according to the degree of toxemia and reflected the general condition of the patient. It reached a peak of 41.5 C. (106.7 F.) on the ninth day, gradually returned to normal on the fifteenth day and remained within normal limits thereafter.

The variation of the pulse coincided almost exactly with that of the temperature. It is worthy of note that the pulse was slow in relation to the degree of fever for the first seven days, during which time the patient exhibited other signs symptomatic of intercerebral pressure, namely, headache, vomiting, mental confusion and stupor. Reference to chart 2 shows that for the first seven days the pressure at the lumbar level, with the patient in the horizontal position, varied between 350 and 280 mm. of water, with the cell count elevated as high as 15,000 per cubic millimeter. Coincidently with a marked improvement in the general condition and rapid fall toward normal for the spinal fluid pressure and cell count, the pulse rate became proportional; its curve crossed the temperature line, and it remained slightly faster during convalescence.

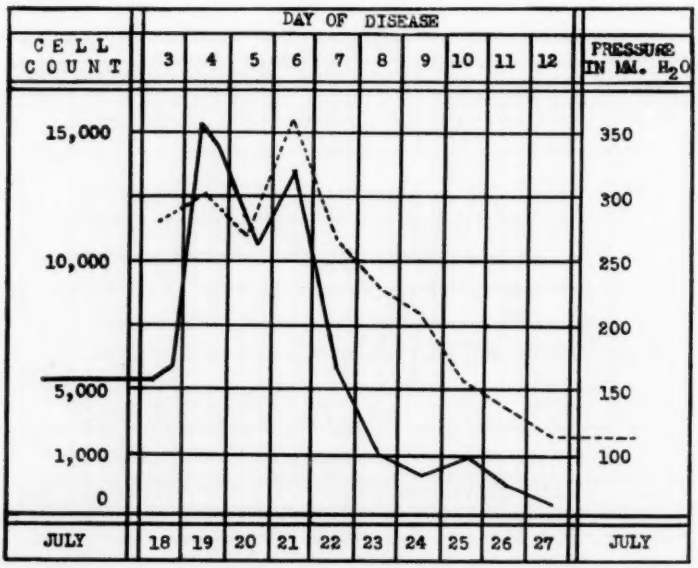


Chart 2.—Data on spinal fluid.

Laboratory Observations.—The urine was normal in all respects. The erythrocyte count and hemoglobin concentration were within the accepted limits. The Wassermann and the Kahn test were negative. The leukocytes ranged between 18,000 and 20,000 per cubic millimeter the first eight days, with a decided shift to the left in the Schilling index. On the fifth day 81 of the 96 per cent granulocytes were nonfilamented forms or younger. The granulocytes predominated throughout the disease. The spinal fluid pressure and cell count fell toward normal just as the Schilling index shifted back to the right, and the total leukocyte count dropped below 10,000 per cubic millimeter. The reaction for globulin remained 4 plus to Pandy's solution through the ninth day.

Cutaneous Reactions.—Early in the disease the patient was tested intradermally with soluble meningococcus toxin with the dilutions and on the dates outlined in table 1. The skin test dose was 0.05 cc. intracutaneously.[2]

The patient was tested by means of intracutaneous wheals to determine his sensitivity to the meningococcus toxin. It was found he reacted even with the dilution of 1:100,000. After the administration of the antimeningococcus serum throughout

TABLE 1.—*Intradermal Tests with Soluble Meningococcus Toxin*

Dilutions of Toxin	Dates Administered, 1936		
	July 17 1st Day of Disease	July 21 4th Day of Disease	July 23 8th Day of Disease
1:100,000	+ — (—)	Negative	Negative
1:10,000	+ — (—)	Negative	Negative
1:1,000	+ — (+)	Negative	Negative
1:100	+	+ —	Negative
1:10	++	+	+ —
Full strength	+++	++	+

TABLE 2.—*Analysis of the Eight Reported Cases of Meningitis Due to M. Tetragenus*

Case No.	Age	Sex	Onset	Duration, Wk.	Outcome	Early Manifestation	Complications and Sequelae	Predisposing Factor
1	32	F	Not given	Not given	Death	Puerperal infection	Septicemia	Childbirth
2	35	M	Sudden	3	Death	None described	None described	None described
3	48	F	Sudden	5	Death	None described	None described	None described
4	?	M	Sudden	2	Recovery	Sudden headache and general malaise	Pulmonary infection	Disease of the upper respiratory tract
5	23	M	Sudden	1	Death	Sore throat and malaise	Septicemia and arthritis	Disease of the upper respiratory tract
6	28	M	Sudden	4	Recovery	Believed to be brucellosis	None described	None described
7	46	M	Sudden	2	Recovery	Painful joints and sore throat	Pharyngitis, arthritis and septicemia	Drunken debauch and disease of the upper respiratory tract
8	20	M	Sudden	2	Recovery	Pain in neck and general malaise	Serum sickness	Heat exhaustion

the disease it was found that he reacted only weakly to the strongest concentrations—plus-minus to 1:10 on July 23, 1936, the eighth day of the disease. The sole significance of this is that immunity to the soluble toxin of the meningococcus may create an immunity to the soluble toxin of M. tetragenus also, since the decrease in cutaneous reactivity did parallel recovery from the disease.

2. Kuhns, D. M.: Control of Meningococcic Meningitis Epidemics by Active Immunization with Meningococcus Soluble Toxin, J. A. M. A. **107**:5 (July 4) 1936.

COMPARISON OF EIGHT CASES OF MENINGITIS DUE TO M. TETRAGENUS

The following cases of meningitis due to M. tetragenous have been reported:

Case Number	Source
1	Benzancon and Lepage [3]
2	Grieve, Fackler and Mitchell [4]
3	Pende [5]
4	Vincent [6]
5	Leschke [7]
6	Bonanno [8]
7	Reimann [1a]

In comparing the clinical features of this case with those of 7 reported cases (table 2), we see that in all the condition had a sudden onset and that the shortest duration was one week and the longest five weeks. Four of the patients made an uncomplicated neurologic recovery, which is in decided contrast to the residuum which meningococcic meningitis so often leaves.

A much larger series of cases will be necessary before statistical significance can be placed on the sex or age distribution. In this small series the sex ratio showed male predominance by 3 to 1. Strangely enough, all attacks were in the virile adult decades, leaving untouched the debilitated aged and the immature. For an organism looked on as nonpathogenic or feebly virulent at most, such an age incidence cannot be explained satisfactorily with the present small series. Perhaps the distribution depends solely on the presence of a conditioning or predisposing factor which has made the subject unusually susceptible. This factor has been reported in a majority of all the cases. In the present case debilitating humid temperature continuing over a long period so lowered the patient's resistance that the organism was able to invade the meninges.

---

3. Benzançon, F., and Lepage: Meningitis Due to Micrococcus Tetragenus, Semaine méd. **18**:40, 1898.

4. Grieve, J. W.; Fackler, G. A., and Mitchell, E. W.: Case of Micrococcus Tetragenus Meningitis, Philadelphia M. J. **1**:528, 1898.

5. Pende, N.: Meningitis of Micrococcus Tetragenus, Policlinico (sez. prat.) **14**:26, 1907; abstracted, Centralbl. f. Bakt. **41**:294, 1908.

6. Vincent, M. H.: Meningitis of Micrococcus Tetragenus, Bull. Soc. med. de Paris **3**:26, 1908.

7. Leschke, E., in Kraus, F., and Brugsch, T.: Spezielle Pathologie und Therapie innerer Krankheiten, Berlin, Urban & Schwarzenberg, 1919, vol. 2, p. 1103; cited by Bonanno [8] and by Reimann.[1a]

8. Bonanno, A. M.: Meningitis Caused by Gaffkya Tardissima, abstracted, Riforma med. **47**:363 (March 9) 1931.

With the foregoing statements in mind it will be instructive to examine in detail tables 3 and 4, which compare meningococcic meningitis with that due to M. tetragenus and to note how few are the differences in their clinical pictures.

TABLE 3.—*Comparison of Symptoms, Physical Findings and Complications of Meningitis Due to M. Tetragenus and Meningococcic Meningitis*

Symptoms and Physical Findings	Meningitis Due to M. Tetragenus	Meningococcic Meningitis
Pharyngitis	+	+
Tonsillitis	+	− +
Sinusitis	+	+
Otitis media	− +	+
Fever	+	+
Rapid pulse	+	+
Headache	+	+
Chills	+	+
Vomiting	+	+
Diarrhea	+	+
Constipation	+	+
Pains in the neck	+	+
Backache	+	+
General malaise	+	+
Pains in the joints	+	+
Hyperactive reflexes	+	+
Kernig sign	+ −	+
Brudzinski sign	+ −	+
Hyperesthesia	+	+
Pupillary inequality	+ −	+
Photophobia	+	+
Insomnia	+	+
Delirium	+	+
Coma	+	+
Petechiae	+	+
Herpes	−	·
Epididymitis	−	+
Peritonitis	−	+

TABLE 4.—*Comparison of Clinical Pathologic Characteristics of Meningitis Due to M. Tetragenus and Meningococcic Meningitis*

	Meningitis Due to M. Tetragenus	Meningococcic Meningitis
Blood culture	Positive—28%	Positive—5 to 80%
Pressure of spinal fluid	Increased	Not always increased
Cell count of spinal fluid	Increased—number up to 20,000	Increased—number up to 90,000
Globulin content of spinal fluid	Increased—++++	Increased—++++
Sugar content of spinal fluid	Decreased	Decreased
Reaction to colloidal gold test	Typical meningitic curve	0000244444
Differential count (Schilling)	Polynucleosis—many young forms	Polynucleosis—many young forms
Direct smear	Extracellular cocci, some gram-positive and some gram-negative	Intracellular and extracellular gram-negative diplococci
Culture of spinal fluid	Rapid growth on almost all mediums	Slow growth on favorable mediums

This comparison proves that the proper diagnosis depends almost entirely on the identification of an organism which on direct smear may be atypical and easily confused with the meningococcus or the staphylococcus. Accordingly, cultural studies to bring out the peculiarities of the organism are essential.

BACTERIOLOGY

Dissociation of M. Tetragenus Isolated in the Present Case.—Reimann [1] pointed out in 1934 that the M. tetragenus which he isolated in a case of meningitis had the tendency to dissociate into three types of colonies, namely, (*a*) yellow, (*b*) large and small white and (*c*) translucent. The yellow colonies produced a large tetrad, the white colonies small cocci, single and in clusters, and the translucent colonies pleomorphic metachromatic cocci. The type found in the translucent colonies was similar to that observed in direct smears from living tissue.

On transfer of the original broth culture to blood agar, the first growth, observed in eighteen hours, consisted of small translucent and white colonies. At the end of forty-eight hours these colonies had differentiated themselves into four types described by Reimann: (*a*) large yellow colonies with a zone of clear hemolysis, (*b*) large white colonies with a zone of green hemolysis, (*c*) small white colonies with a zone of green hemolysis and (*d*) translucent colonies, occasionally medium sized, with a zone of clear hemolysis.

It was noted that once the dissociation was made on artificial mediums, the characteristics of the colonies remained constant. The yellow colony remained yellow, with the exception that minute white daughter colonies appeared at its margin; the white colony on transfer remained smooth, raised, glistening and white. The small white colony, on transfer to plain agar, remained small for three or four days and then turned slightly cream colored. The translucent colony was noted on the original transfer, but we were unable to recover it, although an occasional colony was noted among other types, which on transfer would not remain constant.

Morphology.—The structure of the bacteria composing the different types of colonies was in agreement with the observation made by Reimann. The large yellow colonies were composed of large, deep-staining, gram-positive tetrads and showed the typical tetrad formation.

The large white colonies were composed of deep-staining, all gram-positive cocci and diplococci, small and with no tetrad formation. There was a suggestion of bipolar arrangement. These colonies would have been difficult to distinguish from staphylococci.

The small white colonies were composed of gram-positive and gram-negative cocci. Their structure was as indefinite as their Gram staining.

There were globular forms, diplococcic and coccic forms and metachromatic chained cocci.

The transluent colonies, in our short observation of this type of growth were made up of the same pleomorphic type of cocci.

Cultural Characteristics on Broth Mediums.—The yellow colony retained its color in broth mediums. The pellicle was yellow and more prolific than that formed by the other colonies. In 500 cc. of beef

TABLE 5.—*Cultural Characteristics of M. Tetragenus* *

Type of Colony on Agar Plate	Structure of Cocci	Growth on Broth	Growth on Gelatin	Growth on Blood Plate	Dextrose	Lactose	Mannite	Xylose	Maltose	Sucrose	Dulcitol
Yellow colony, elevated, lobate edges	Micrococci large, gram-positive, dividing in two planes	Yellow pellicle dropping to form weedy sediment	No liquefaction	Yellow colony with clear zone of hemolysis	+ 3d day	—	—	—	+ 3d day	—	—
White colony, large, smooth elevated, dome shaped	Cocci and diplococci, all gram-positive, smaller, with no tetrads	White pellicle later forming weedy glutinous sediment	Slow liquefaction of gelatin	White colony with green zone of hemolysis	+ 1st day	+	—	—	+ 1st day	+ 2d day	—
White colony, small, convex	Gram-positive and gram-negative cocci and diplococci with globular forms	White pellicle with granular sediment	Slow liquefaction of surface	Early clear hemolysis, later green	+ 1st day	—	—	—	+ 1st day	—	—
Translucent colony, flat, clear, large and small	Same as small white colony	Fragile pellicle with clear underlying liquid, later drops	.....	Clear zone of hemolysis	Not done						
Whitish colony, glutinous, difficult to emulsify	Gram-positive, spherical dividing in two planes, with tetrads	Supernatant liquid, clear; weedy spiraling deposits	.....	.....	+	+	—	—	+	+	—
					High pathogenicity for mouse						

* Acetic acid odor for yellow and white colonies.

infusion broth this pellicle would form completely in three days and disintegrate on the fourth day. In addition to the yellow color, there was a greenish discoloration, which is characteristic of the proteose peptone mediums.

Cultures of the large and of the small white colonies were similar on broth mediums, in that they maintained their white color in broth, formed a pellicle and on its disintegration formed the same glutinous thready sediment.

The yellow colonies did not liquefy gelatin, but the large and small white colonies did. This was one of the few means of differentiating the organism from the staphylococcus.

Sugar Reactions.—The yellow colonies did not form acid on dextrose and maltose until the third day, while the large and small white colonies formed acid within twenty-four hours. The large white colonies did not ferment sucrose until the second day or lactose until the third.

For all cultures a distinct odor was noted, not unlike that of vinegar or acetic acid.

Pathogenicity for Animals.—An agar slant culture of each of the three types of colony, suspended in saline solution, was inoculated intravenously into a rabbit and a guinea pig. This was done to determine if one type of colony was more virulent or produced more toxin than the other. There was no pathogenic effect whatsoever from these inoculations. None of these types killed mice.

Immunity.—The patient was given polyvalent antimeningococcus serum throughout the course of the disease and recovered.

TABLE 6.—*Agglutination Tests of Three of M. Tetragenus Types of Colony Against Gordon's Four Types of Antimeningococcus Serum*

Type of Colony	Gordon's Four Types of Antimeningococcus Serum			
	Type 1	Type 2	Type 3	Type 4
Yellow colony	1:10	1:80	1:20	1:20
Large white colony	Negative	Negative	Negative	Negative
Small white colony	Negative	Negative	Negative	Negative

Early in the disease it was found that M. tetragenus was agglutinated by polyvalent antimeningococcus serum with dilutions as high as 1 : 160. Therefore, further agglutination tests were carried out against the four types of meningococcus to determine if this organism held any relation to Gordon's four types.

From the results of the agglutination tests as seen in table 6, it would appear that, serologically, the yellow colony was most nearly related to the meningococcus. Therefore, if the production of an antiserum for this infection is to be considered, the yellow colony would probably be of the greatest value as an antigen. The serum of the patient during the disease and after recovery did not agglutinate the meningococcus or any of the three types of colony of M. tetragenus.

SUMMARY

A case of meningitis due to M. tetragenus has been reported.

The clinical features, symptoms and laboratory findings were similar to those of meningococcic meningitis. The only differential point was the identification of an organism, atypical in the original smears, but becoming typical of M. tetragenus tetrads in broth cultures.

Microbic dissociation was found. The yellow colony was the undissociated type. It formed daughter white colonies which differentiated into large and small white colonies. The typical structure of the organism was found in the large yellow colonies in the form of tetrads. The large white colonies furnished only cocci and diplococci, which were difficult to distinguish from staphylococci. The small white colony was still further dissociated, in that no tetrads were seen, and the form of the cocci was indeterminate.

A table presenting the cultural characteristics of the three types of colony has been presented. It was found that, once dissociated, each type of colony on transfer to various types of medium maintained its characteristics.

Agglutination tests—the three types of colony against the patient's own serum and Gordon's four types of antimeningococcus serum—were carried out. The yellow colony was not agglutinated by the patient's own serum but was agglutinated by antimeningococcus serum, type 2, with a dilution of 1:80.

The three types of colony were cultured individually in 500 cc. of proteose hormone broth and within four days filtered through a Pasteur-Chamberland filter. The filtrates were tested on three types of animal, rabbit, guinea pig and mouse. Cutaneous tests were made with the filtrates in strength varying from 1:100,000 dilution to full strength on 10 persons.

CONCLUSIONS

1. It has been observed that M. tetragenus is not the harmless organism commonly supposed but under certain circumstances is pathogenic.

2. It occurs as an etiologic agent much more frequently than is recognized, usually being mistaken for a meningococcus or a staphylococcus.

3. Its characteristics of dissociation should be recognized in order to establish its identity.

4. It elaborates a soluble toxin which produces a reaction in the skin of human beings but is not demonstrably toxic for animals.

EXPERIMENTAL PRODUCTION OF NEUTROPENIA WITH AMINOPYRINE

E. M. BUTT, M.D.

A. M. HOFFMAN, M.D.
AND
S. N. SOLL, M.D.
LOS ANGELES

The reproduction of the classic clinical picture of malignant neutropenia, or agranulocytic angina, in experimental animals has never been accomplished successfully. The situation in regard to aminopyrine as the most common etiologic agent in agranulocytic angina in man is much the same today as that in regard to cinchophen as the cause of acute necrosis of the liver. Cinchophen is generally accepted as the toxic agent in production of acute yellow atrophy in certain susceptible persons. In the experimental animal, however, cinchophen has never produced necrosis of the liver in any degree sufficient to simulate the condition in man. Aminopyrine today is accepted by many as the responsible agent in agranulocytic angina. True, it is apparently harmless to the vast majority of patients. Rawls [1] estimated on clinical grounds that only 1 to 2 per cent of patients are susceptible to demonstrable changes in the peripheral blood; this view is based on an "individual susceptibility." If the same numerical proportion exists in animals, it is understandable that the experimental reproduction of the disease would be difficult, if only because of the large number of animals required. It is our belief on the basis of our efforts during the past three years to reproduce the disease with aminopyrine in animals that the toxic action of the drug is far more frequent than Rawls' figures would indicate. We believe that changes in the bone marrow occur in the majority of animals but that changes in the peripheral blood are seen in only a small proportion.

Attempts at experimental reproduction of neutropenia have been numerous. In 1932, before the attention of workers was focused on

From the Departments of Pathology and Medicine, University of Southern California School of Medicine, and the Santa Fe Coast Lines Hospital.

1. Rawls, W. B.: Effect of Amidopyrin on Red, White, and Polymorphonuclear Blood Cells of a Series of One Hundred Patients, Am. J. M. Sc. **192**:175 (Aug.) 1936.

aminopyrine, Kracke [2] reported successful reproduction of agranulocytosis in rabbits with small subcutaneous or intraperitoneal doses of benzene. Large doses administered by the same technic affected the erythroblastic elements as well. Intravenous injection of benzene killed the rabbits immediately. Marked leukopenia was produced by the subcutaneous injection of salicylic acid and by intravenous administration of hydroquinone. Oral, subcutaneous or intravenous use of aminopyrine, acetophenetidin, aminopyrine diethylbarbiturate, dial, resorcinol, pyrocatechin, orthocresol, metacresol, paracresol, phenol, paraoxybenzoic acid, metaoxybenzoic acid and 50 per cent alcohol failed to affect the leukocyte count.

Fried and Dameshek,[3] in 1932, infected rabbits intravenously with Salmonella suipestifer and caused temporary neutropenia. Kracke [4] justly pointed out that such temporary neutropenia can be produced "by a wide variety of substances, including milk, nonspecific proteins, dead and living bacteria and various types of inert, finely divided material" and that such temporary neutropenia in no way simulates true agranulocytosis.

Dennis [5] placed cultures of Staphylococcus aureus, Streptococcus haemolyticus, Streptococcus viridans and Proteus septicus in parchment capsules, inserted the capsules into the abdominal cavities of rabbits and observed what he interpreted as granulopenia. Most of the rabbits showed no drop, but rather a rise, in the total leukocyte count, while some showed a moderate to marked decrease in the polymorphonuclear count.

Meyer and Thewlis [6] were unable to obtain results similar to those of Dennis. With Bacillus pyocyaneus, however, they produced leukopenia in rabbits, with death in forty hours but with no decrease in the number of polymorphonuclear cells. Ricci [7] inoculated rats with living cultures of S. suipestifer and B. pyocyaneus and produced varying degrees of

2. Kracke, R. R.: The Experimental Production of Agranulocytosis, Am. J. Clin. Path. **2**:11 (Jan.) 1932.

3. Fried, B. M., and Dameshek, W.: Experimental Agranulocytosis: Infections of Rabbits with Salmonella Suipestifer by Way of the Blood Stream, Arch. Int. Med. **49**:94 (Jan.) 1932.

4. Kracke, R. R.: "Experimental Agranulocytosis," Arch. Int. Med. **49**:709 (April) 1932.

5. Dennis, E. W.: Experimental Granulopenia Due to Bacterial Toxins Elaborated in Vivo, J. Exper. Med. **57**:993 (June) 1933.

6. Meyer, O. O., and Thewlis, E. W.: A Report of Failure to Produce Granulocytopenia with Bacterial Toxins, J. Clin. Investigation **13**:437 (May) 1934.

7. Ricci, F. C.: Sulla agranulocitosi sperimentale, Arch. ital. di otol. **46**:401 (June) 1934.

leukopenia and agranulocytosis. In his animals, however, abscesses in the liver developed. Harris and Schattenberg [8] claimed that by injecting prepared toxins of Bacillus enteritidis and Str. haemolyticus into guinea pigs they had reproduced the blood picture found in human beings. Staph. aureus from a patient who had died of the disease brought about depression of the total leukocyte count and neutropenia in guinea pigs but not in rabbits.

De Vries' [9] first indication, in September 1933, of the possible relation of aminopyrine to granulopenia in human beings, followed by the reports of Madison and Squier,[10] Watkins,[11] Hoffman, Butt and Hickey,[12] Randall [13] and many others, gave a great impetus to the experimental study of the disease, which was now centered about aminopyrine as the etiologic agent. The majority of the authors reported satisfactory results. Strangely enough, the earliest of their observations may be one of the most significant. In a short note, Miller [14] reported results of oral administration of 0.3 Gm. of a 5 per cent solution of aminopyrine per kilogram of body weight to 16 dogs. It was given for four weeks, and no instance of depression of the total number of circulating leukocytes was encountered.

Histological study of the femoral bone marrows of the treated animals, however, revealed striking changes, which gave evidence that the drug does affect the formation of granulocytes, even though not to a degree which is reflected in a materially decreased number of granular leukocytes in the circulating blood. A well-defined suppression of maturation of the hematopoietic elements occurs.

---

8. Harris, W. H., and Schattenberg, H. J.: Experimental Studies in So-Called Agranulocytic Angina: I. Effects upon Leucocytes by Toxic Products of Bacteria from Stool of Human Case, Proc. Soc. Exper. Biol. & Med. **31**:843 (April) 1934. Schattenberg, H. J., and Harris, W. H.: II. Effects upon Leucocytes by Toxic Products of Staphylococcus Aureus from Blood of Human Case, ibid. **31**:847 (April) 1934.

9. de Vries, S. I., Jr.: Recurrent Agranulocytic Syndrome, Nederl. tijdschr. v. geneesk. **77**:4443 (Sept. 30) 1933.

10. Madison, F. W., and Squier, T. L.: (*a*) Primary Granulocytopenia After Administration of Benzene Chain Derivatives, J. A. M. A. **101**:2076 (Dec. 23) 1933; (*b*) The Etiology of Primary Granulocytopenia, ibid. **102**:755 (March 10) 1934.

11. Watkins, C. H.: The Possible Role of Barbiturates and Amidopyrine in the Causation of Leucopenic States, Proc. Staff Meet., Mayo Clin. **8**:713 (Nov. 22) 1933.

12. Hoffman, A. M.; Butt, E. M., and Hickey, N. G.: Neutropenia Following Amidopyrine, J. A. M. A. **102**:1213 (April 14) 1934.

13. Randall, C. L.: Severe Granulopenia Following the Use of Barbiturates and Amidopyrine: Report of a Case, J. A. M. A. **102**:1137 (April 7) 1934.

14. Miller, D. K.: Histological Changes in the Bone Marrow of the Dog Following Amidopyrine Administration, Science **80**:320 (Oct. 5) 1934.

Granulocytes are decreased in numbers or almost completely absent. The number of young, relatively undifferentiated cells is increased. Many erythroblasts and myeloblasts are present, as are cells of an even more primitive type; more adult forms are rare. In certain more advanced cases the orderly arrangement of the bone marrow structure into hematopoietic islands has been disturbed.

From the observations reported it appears that amidopyrine orally administered may exert a toxic effect upon the bone marrow, with little or no evidence of the fact in the circulating granular elements of the blood.

These observations are consistent with those on human bone marrow in fatal cases reported by Fitz-Hugh and Krumbhaar [15] in a paper in which they proposed the hypothesis of "maturation arrest." Custer [16] reported observations which he made on human bone marrow in 11 cases, using marrow from the tibial and femoral shafts as an example of that from the long bones and marrow from the vertebrae, sternum or ribs as an example of that from cancellous bone. He pointed out that an examination based on marrow removed from but one locus is "woefully inadequate." In 9 of the 11 cases of proved idiopathic agranulocytosis he observed:

(*a*) *Marked proliferation of myeloblasts.*

(*b*) Failure of these cells to mature, resulting in paucity of myelocytes and practically complete absence of segmented forms.

(*c*) Normal or slightly increased formation of red blood cells.

(*d*) Slight hyperplasia of otherwise normal megakaryocytes.

(*e*) Infiltration of lymphocytes and plasmocytes.

The careful observations both of Fitz-Hugh and Krumbhaar and of Custer were amply confirmed by Darling, Parker and Jackson [17] in a study of the bone marrow of 24 persons who died and of the sternal bone marrow of 1 patient who recovered. They suggested the term "granulocytic anakmesis" for the type of arrest of maturation observed in the bone marrow in agranulocytic angina.

Madison and Squier,[10b] in their clinical report, described their attempts to produce agranulocytosis in rabbits. Eleven rabbits received allonal (allylisopropylbarbituric acid with aminopyrine) by mouth in doses of 2⅔ to 24 grains (0.17 to 1.55 Gm.) daily. One rabbit which had received an average of 20 grains (1.29 Gm.) daily

showed a rather abrupt drop in both the total and the granulocyte count on the twenty-fifth day, following which there was a progressive fall until death on

15. Fitz-Hugh, T., Jr., and Krumbhaar, E. B.: Myeloid Cell Hyperplasia of the Bone Marrow in Agranulocytic Angina, Am. J. M. Sc. **183**:104 (Jan.) 1932.

16. Custer, R. P.: Studies on the Structure and Function of the Bone Marrow: IV. Bone Marrow in Agranulocytosis, Am. J. M. Sc. **189**:507 (April) 1935.

17. Darling, R. C.; Parker, F., and Jackson, H.: The Pathological Changes in the Bone Marrow in Agranulocytosis, Am. J. Path. **12**:1 (Jan.) 1936.

the thirtieth day. . . . Preceding death there was complete absence of granulocytes in the peripheral blood and the blood picture was characteristic of primary granulocytopenia except for a moderate anemia. At necropsy there were no grossly abnormal conditions found save that the bone marrow was absolutely lacking in cells of the granular series. None of the other rabbits showed the blood picture of granulocytopenia.

No mention was made, however, of observations on the bone marrow of these rabbits.

In our early report [12] we described our results with oral administration of aminopyrine to rabbits in doses of 0.2 to 0.9 Gm. per kilogram. Definite leukocytosis was produced in all the animals, followed in a few weeks by a depression of the total white cell count. In all the rabbits the polymorphonuclear leukocyte count was reduced to 20 per cent, while a few animals showed depression to as low as 8 per cent. However, because of the known wide normal variation of the leukocyte counts of rabbits, we felt that these results were not conclusive and therefore limited further experimental study to dogs.

Stenn [18] reported observations on the effect of aminopyrine on 120 animals, including the guinea pig, rabbit and monkey, over a period of eight months and concluded: "Administration of amidopyrine . . . both with and without preliminary anemia or bone marrow injury, failed to produce any appreciable granulocytopenia." No observations on the bone marrow were recorded.

Climenko [19] observed that after oral administration of aminopyrine to rabbits—without depression of granulocytes in the circulating blood—the leukocytosis which usually follows injection of nucleic acid intramuscularly failed to occur. The bone marrow of the experimental animals showed marked hyperplasia, with an increase in the number of primitive hemocytoblasts. Essentially the same type of inhibition of the bone marrow preceded by an increase in the percentage of juvenile forms without concomitant leukocytosis was produced after administration of α-dinitrophenol. The first case of agranulocytic angina due to α-dinitrophenol in man was reported in our original paper.[12]

Kunde, Herwick, Learner and Sternback [20] administered a preparation of aminopyrine and dial (cibalgin) to 35 rabbits with endemic

18. Stenn, F.: Etiologic Relationship of Amidopyrine to Agranulocytosis, J. Lab. & Clin. Med. **20**:1150 (Aug.) 1935.

19. Climenko, D. R.: Inhibition of Leucogenic Activity in the Rabbit by Certain Cyclic Compounds, Proc. Soc. Exper. Biol. & Med. **32**:823 (March) 1935.

20. Kunde, M. M.; Herwick, R. P.; Learner, A., and Sternback, M.: Non-Production of Granulocytopenia with an Amidopyrine Compound in Some Acute Infections, Proc. Soc. Exper. Biol. & Med. **32**:1121 (April) 1935.

laboratory infections, such as sniffles and acute gastrointestinal disorders. They concluded that the drug did not diminish the number of leukocytes in normal rabbits when given for seventeen days and observed no depression of the leukocytosis present in the infected animals. Thyroidectomized animals showed the same results as the others. No observations on the bone marrow are recorded.

Bolton [21] administered 0.3 Gm. of aminopyrine in water by stomach tube to a dog over a period of twenty-three days. The leukocyte count remained elevated from 18,000 to 20,000, with no significant change in the differential count. Sections from the femur, humerus and ribs showed "hyperplasia of the bone marrow, with no decrease in the myeloid elements." In another dog (experiment 11) larger doses of aminopyrine were followed by depression of the original leukocyte count of 12,200 to 6,500. There was a subsequent rise to the original level, with no change in the percentage of granulocytes. Additional doses were given intravenously—altogether, 4.5 Gm. of the drug for each kilogram of body weight over a period of thirty-three days. No necropsy was reported.

Smith and Mack [22] studied the effect of amytal and aminopyrine given orally to albino rats fed a deficient diet. They concluded that the total white cell count could be reduced approximately 50 per cent in the animals weakened by the deficient diet, but they did not note any change in granulocytes or any abnormality in the bone marrow.

Wilson [23] administered phenobarbital and aminopyrine subcutaneously, both in combination and separately, to white rats for twenty-eight days and noted only leukocytosis. A few normoblasts were found in the aminopyrine series, and he concluded that the bone marrow was affected in some manner. The bone marrow was not studied.

Miller and Rhoades [24] reported further work on the effect of a deficient diet combined with aminopyrine on dogs. Dogs fed 0.5 Gm.

---

21. Bolton, V. L.: Laboratory Study of Amidopyrine, Barbital, Phenyl Hydrazine, and Benzene in Relation to Agranulocytic Angina, J. Lab. & Clin. Med. **20**:1199 (Aug.) 1935.

22. Smith, E., and Mack, L.: Effect of a Deficient Diet, Amytal and Amidopyrine on the Blood Picture of the Albino Rat, Proc. Soc. Exper. Biol. & Med. **32**:1623 (June) 1935.

23. Wilson, S. J.: Effects of Amidopyrine and Phenobarbital on the Blood Cell Count of White Rats, J. Kansas M. Soc. **36**:500 (Dec.) 1935.

24. Rhoades, C. P., and Miller, D. K.: Effect of Diet on the Susceptibility of the Canine Hematopoietic System to Damage by Amidopyrine, Proc. Soc. Exper. Biol. & Med. **36**:654 (June) 1937. Miller, D. K., and Rhoades, C. P.: Effect of Diet on the Susceptibility of the Canine Hematopoietic Function to Damage by Amidopyrine, J. Exper. Med. **66**:367 (Sept.) 1937.

of aminopyrine daily failed to demonstrate a change in the blood picture. They further found that in dogs receiving the Goldberger pellagra-producing (blacktongue) diet appreciable anemia usually failed to develop. The combination of the drug and the diet, however, produced pronounced and often fatal anemia in from eight to thirty-five days. There was remarkable absence of leukopenia. The bone marrow histologically resembled that described in cases of benzene poisoning.

Numerous experimental observations on man have been recorded. Benjamin and Biederman,[25] Kloster,[26] Hansen and Holten,[27] Plum [28] and Dameshek and Colmes [29] have each reported on from 1 to several patients with known susceptibility to aminopyrine who had granulopenia again when aminopyrine was administered after recovery from the original attack. Hourly counts were frequently significantly altered immediately after ingestion of the drug. Most of the patients so tested gave negative responses to aminopyrine in intradermal scratch, passive transfer and patch tests. Dameshek and Colmes, however, attempted intradermal tests with blood serum previously "sensitized" by aminopyrine. These gave positive results in the 3 cases in which they were tried.

EXPERIMENTS

Aminopyrine (dimethylaminoantipyrine) was fed to a group of 7 dogs. An eighth, dog 5 in the series, was kept as a control. The drug was administered to dogs 1, 2 and 3 through a stomach tube. Dogs 4, 6, 7 and 8 in a second series of experiments were fed gelatin capsules containing the powdered drug.

The initial dose of aminopyrine fed to dogs 1, 2 and 3 was approximately 0.5 Gm. per kilogram. Ten minutes after the administration dog 2 died in convulsions. Dogs 1 and 3 vomited a portion of the drug and several days later had completely recovered from the toxic effects of the initial dose. The dose of aminopyrine

25. Benjamin, J. E., and Biederman, J. B.: Agranulocytic Leukopenia: Report of a Case Successfully Treated with X-Rays and Some Observations on the Effect of Amidopyrine, J. A. M. A. **103**:161 (July 21) 1934.

26. Kloster, J.: A Case of Granulocytopenia Produced Experimentally by Pyramidon "Bayer," Acta med. Scandinav., 1936, supp. 78, p. 595.

27. Hansen, A. B., and Holten, C.: Hypersensitiveness to Amidopyrin with Resulting Granulopenic Reaction, Acta med. Scandinav., 1936, supp. 78, p. 599.

28. Plum, P.: Studies on the Etiological Significance of Amidopyrin (Pyramidon) in Agranulocytosis, Acta med. Scandinav., 1936, supp. 78, p. 605; Effect of Amidopyrine on Granulocytopoiesis of Bone Marrow: Continued Investigations on the Etiology of Agranulocytosis; Preliminary Report, Ugesk. f. læger **98**:91 (Jan. 30) 1936.

29. Dameshek, W., and Colmes, A.: Effect of Drugs in the Production of Agranulocytosis with Particular Reference to Amidopyrine Hypersensitivity, J. Clin. Investigation **15**:85 (Jan.) 1936.

was then reduced to 0.05 Gm. per kilogram and was gradually increased to approximately 0.35 Gm. per kilogram. Dog 3 lived for seven months, receiving a total of 136.4 Gm. of the drug, administered in sixty-four doses. On two occasions convulsions developed in this animal and were successfully treated with barbital and caffeine with sodium benzoate. Dog 1 survived for ten months, receiving a total of 250.9 Gm. of aminopyrine, in one hundred and twenty-three doses.

The leukocyte count for dog 3 varied from 8,500 to 12,000. On two occasions leukocytosis (14,500 and 15,500 cells) was noted. The white cell count did not

TABLE 1.—*Details of Experiments on 8 Dogs*

Dog No.	Dose of Aminopyrine, Gm./Kg.			Total Administration		Variation in Blood Count		Changes in the Bone Marrow	Duration of Experiment, Mo.	Comment
	Initial	Lowest	Highest	Gm.	No. of Doses	Leukocytes	Erythrocytes			
1	0.52 (Very ill)	0.055	0.28	250.9	123	45,000 to 350	9,130,000 to 4,900,000	Marked aplasia	10½	Died during stage of marked granulopenia
2	0.54	.....	....	.....	...	.....	.....	.....	....	Died 10 minutes after administration
3	0.53 (Vomited some of drug)	0.058	0.35	136.4	64	15,000 to 6,000	9,100,000 to 3,500,000	Moderate aplasia	6½	Comatose for 12 hr.; died of air emboilsm
4	0.12	.....	0.12	2.0	2	.....	.....	.....	....	Sick at beginning of experiment
5	Control dog	.....	....	.....	...	8,000 to 12,000	Count not made	None	10½	
6	0.06	0.06	0.22	26.75	29	14,000 to 6,000	6,300,000	Slight aplasia	2½	Found dead in cage
7	0.027	0.027	0.21	255.5	167	22,000 to 6,000 (No counts 5 days before death)	7,400,000 to 5,200,000	Marked aplasia	9	Found dead in cage
8	0.036	0.036	0.21	60.2	65	17,500 to 5,500	.....	Slight aplasia	4	Died while passing stomach tube

fall below 6,000. The percentage of polymorphonuclear leukocytes varied from 52 to 80. The red cell count was 9,100,000 at the beginning of the observations and 6,400,000 at the end of the experiment. The platelets were not counted but appeared numerous in all slides.

The white cell count of dog 1 dropped to 900 at the end of the fifth month. The dose of aminopyrine was reduced from 0.28 to 0.14 Gm. per kilogram, and two days later the white cell count increased to 32,000, of which 95 per cent were polymorphonuclear cells. The dose of aminopyrine was then gradually increased to 0.33 Gm. per kilogram, but later it was dropped to 0.25 Gm. per kilogram, as the former dose was too toxic. The day before the death of the animal the white blood cell count was 350. No polymorphonuclear leukocytes were found in the blood smears.

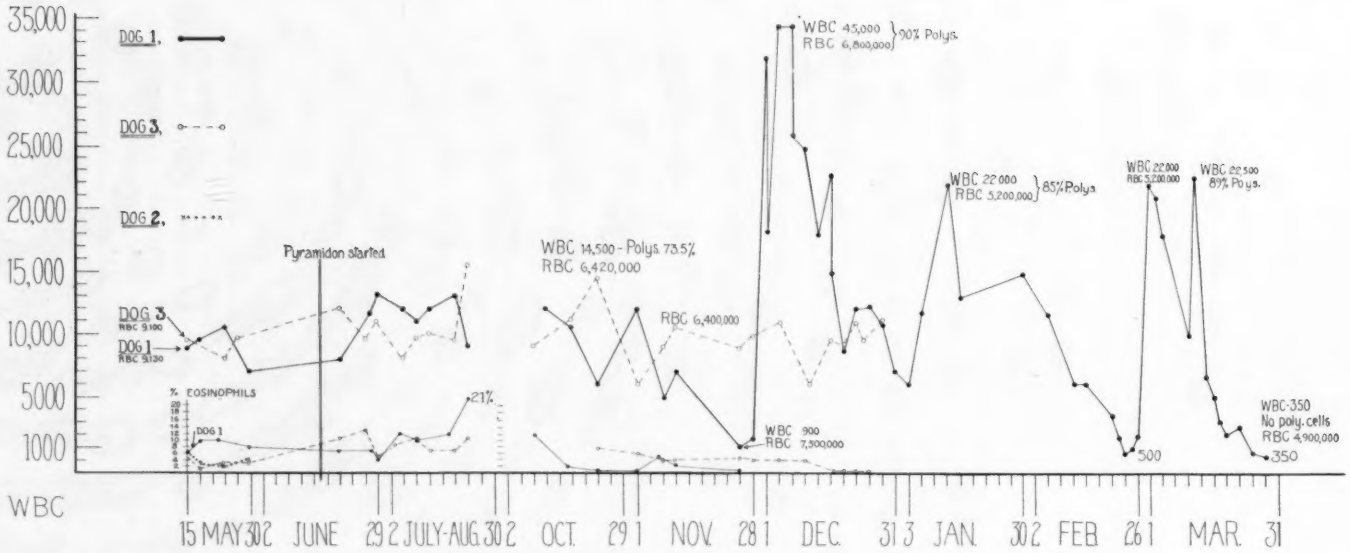


Fig. 1.—Details of experiments on dogs 1, 2 and 3.

Dog No.	Weight During Experiment, Kg. Beginning	End	Gm. of Aminopyrine per Kg. of Body Weight: Initial Dose	Reduced to	Increased to	Total Drug Given, Gm.	Total Number of Doses	Comment
1	9	5.75	0.52	0.055	0.28	250.9	123	
2	...	....	0.54	.....	....	0.54	1	Death in 10 minutes
3	8.5	7.2	0.53	0.058	0.35	136.4	64	Vomited initial dose
6	...	....	0.06	.....	0.22	26.75	29	
7	9.2	6.8	0.027	.....	0.4	255.5	167	
8	...	....	0.036	.....	0.21	60.2	65	

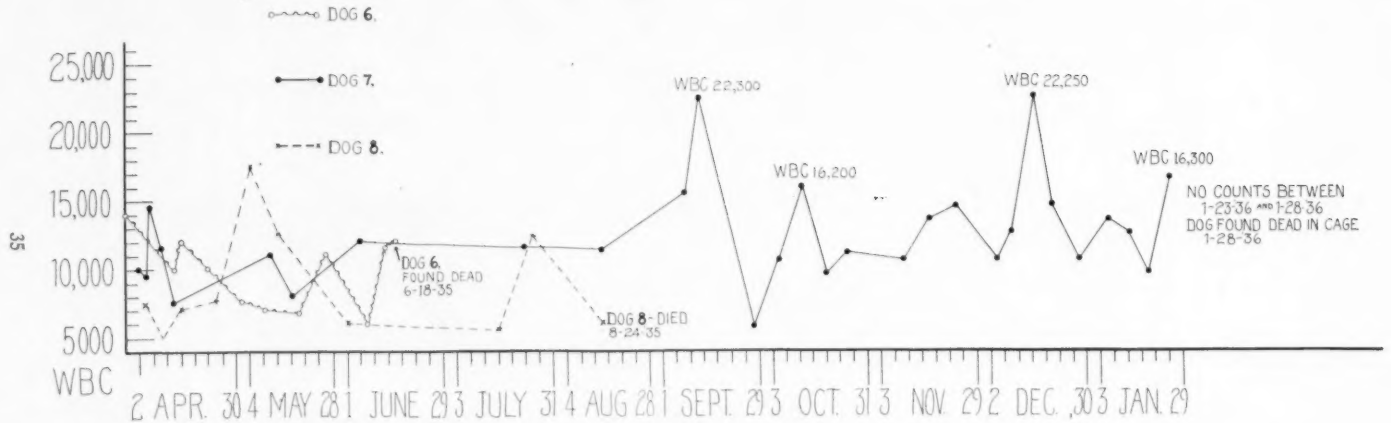


Fig. 2.—Details of experiments on dogs 6, 7 and 8.

Some details of the blood counts of dogs 1 and 3 are recorded in table 2. Other details of the experiments appear in figure 1 and table 1.

Dog 4 died of a pulmonary infection shortly after the beginning of the experiment. Details of the experiments conducted with dogs 6, 7 and 8 are found in figure 2 and table 1. Dog 7 was the only one of this series to survive any great

TABLE 2.—*Details of Blood Counts of Dogs 1 and 3*

Date	Erythrocyte Count	Leukocyte Count	Polymorphonuclear Neutrophilic Leukocytes, %	Lymphocytes, %	Monocytes, %	Eosinophils, %	Basophils, %
Dog 1							
5/15/34	9,130,000	8,900	56	33	5	6	..
5/18/34	7,610,000	9,700	62	23	5	10	..
5/23/34	9,440,000	10,400	60	24	6	10	..
5/29/34	9,200,000	7,200	53	36	3	8	..
6/20/34	9,760,000	8,000	62	24	7	7	..
	(Administration of aminopyrine started)						
6/27/34	7,980,000	11,500	59	30	4	7	..
7/20/34	8,160,000	9,100	59	14	6	21	..
10/23/34	11,000,000	5,600	49	31	18	2	..
11/ 6/34	7,300,000	5,000	60	32	4	4	..
11/26/34	7,500,000	900	2	93	5	..	..
11/27/34	6,800,000	1,250	1	94	5	..	..
12/ 3/34	.....	45,000	90	5	5	..	..
12/20/34	4,820,000	8,300	84	5.5	10.5	..	..
1/29/35	.....	14,700	..	..	..	..	..
2/11/35	.....	6,000	69	22	9	..	..
2/25/35	.....	800	18	76	6	..	..
2/28/35	.....	22,000	54	30	16	..	..
3/16/35	.....	5,000	..	..	..	..	..
3/25/35	.....	400	16	72	12	..	..
3/27/35	.....	350	0	98	2	..	..
3/28/35	Died						
Dog 3							
5/15/34	9,100,000	9,700	69	23	3	5	..
5/18/34	8,200,000	9,150	68	29	2	1	..
5/23/34	8,790,000	8,000	73	17	7	3	..
5/29/34	7,840,000	9,800	51	40	6	3	..
	(Administration of aminopyrine started)						
6/27/34	7,380,000	9,800	54	31	2	13	..
7/ 4/34	6,060,000	7,700	62	21	8	9	..
7/20/34	6,530,000	15,500	68	17	4	11	..
11/ 1/34	.....	6,200	68	17	8	6	1
12/12/34	.....	6,000	81	11	7	1	..
12/24/34	3,500,000	9,400	74	18	7	..	..
12/30/34	Died						

length of time. Unfortunately, no white blood cell counts were made on this dog for five days preceding death. It is probable that the white cell count was low, as the bone marrow was fully as aplastic as that found in dog 1.

The presentation of the main details of the observations at autopsy is limited to animals 1, 3, 7 and 8, as the few changes noted in the remainder of the dogs are not relevant to the problem. None of the dogs had gangrenous or ulcerative lesions of the gastrointestinal tract. Grossly the various organs, except the bone marrow, appeared normal. Microscopically the spleens contained considerable

amounts of coarsely granular, iron-containing pigment. Erythroblasts were observed in the spleens of dogs 1 and 7, and the lymphoid tissue was scanty.

The bone marrow of dogs 3 and 8 could not be distinguished grossly from normal marrow, except perhaps for some increase in red marrow. The bone marrow of dogs 1 and 7 was interesting in that nearly all the marrow was thin

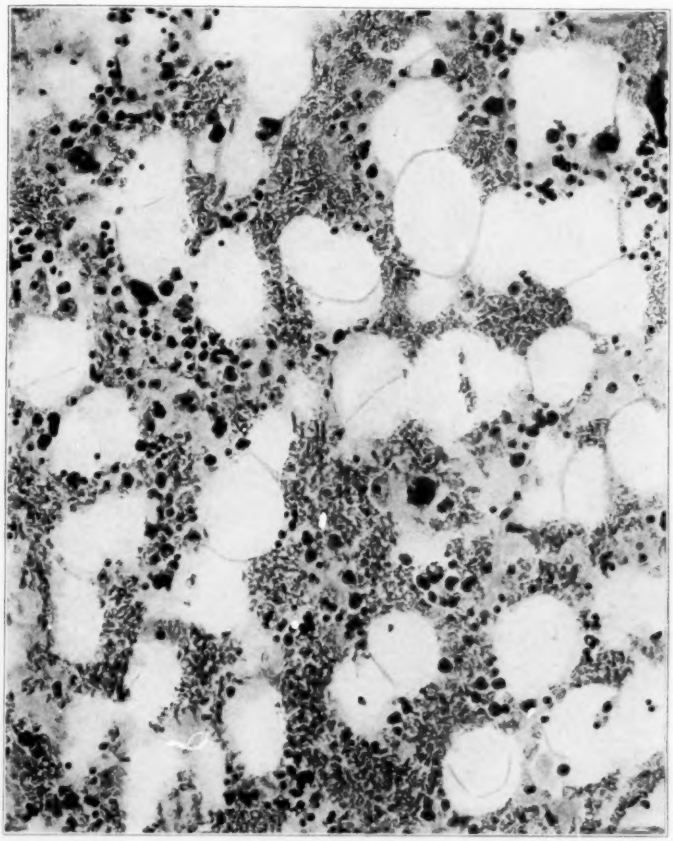


Fig. 3 (dog 1).—Bone marrow of the humerus (× 310).

and red and contained streaks of fat. Histologic examination revealed nearly complete aplasia of the myeloid elements of the marrow. There was noticeable aplasia of the erythroblastic cells. Between the small islands of bone marrow tissue were dilated vascular channels filled with red blood cells and fatty tissue. The megakaryocytes appeared to be reduced in number.

Microscopically the marrow of dog 3 presented moderate but definite aplasia of the myeloid cells, with relative increase of the erythroblastic cells. Minor changes in the marrow were noted in dogs 6 and 8, which could be interpreted as slight hyperplasia or slight aplasia of the myeloid cells, depending on the location of the marrow.

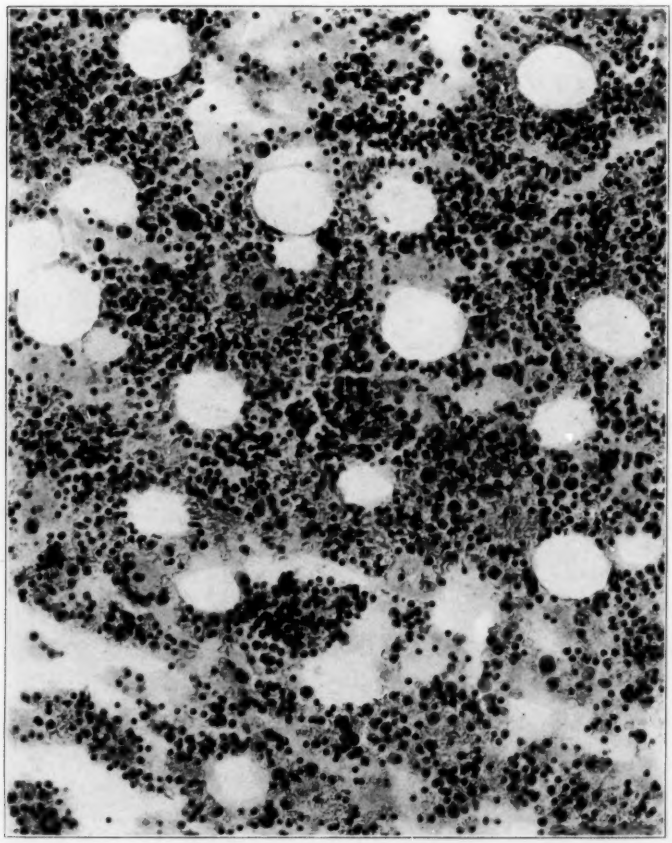


Fig. 4 (dog 3).—Bone marrow of the humerus (× 310).

An interesting relation between the total number of grams of aminopyrine administered and the degree of aplasia of the bone marrow is noted in table 1. The most marked aplasia of the bone marrow was observed in the 3 dogs receiving the largest amounts of aminopyrine. Three of the remaining dogs died of the systemic toxic effects of the drug before changes in the bone marrow were sufficiently marked to be unequivocal.

In the foregoing account it has been clearly demonstrated that aplasia of the bone marrow of dogs follows the oral administration of large quantities of aminopyrine. The action is not unlike that of benzene and is somewhat selective, as the destruction of myeloid elements pre-

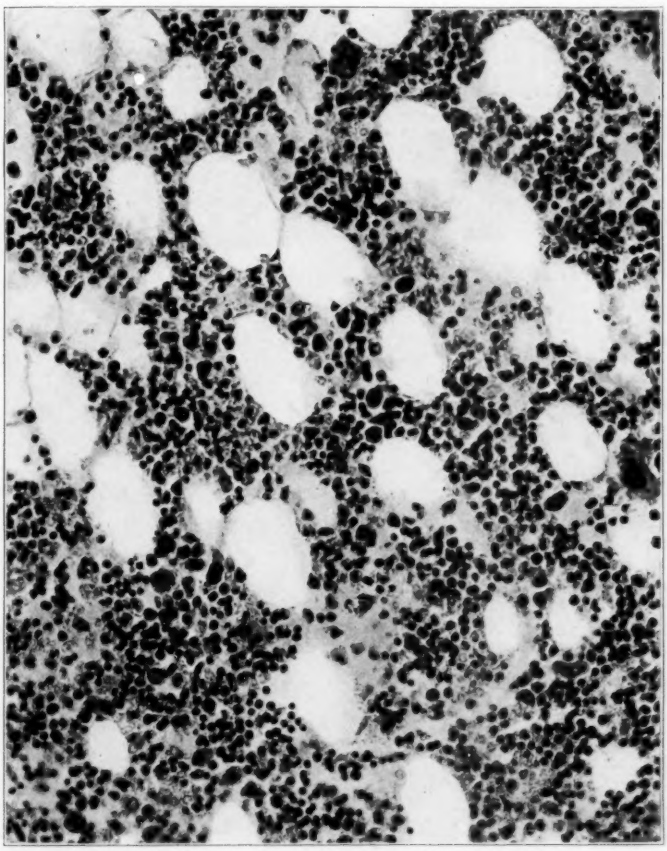


Fig. 5 (dog 6).—Bone marrow (× 310).

cedes the aplasia of the erythroblastic cells. Furthermore, these experiments emphasize the fact that individual variation in susceptibility to the toxic effects of the drug is difficult to evaluate in grams per kilogram. In these experiments, however, we were concerned only with the toxic effects of aminopyrine, and not with individual susceptibility. Such an experimental predication requires that the dose of the drug be graduated

to show its toxic effects rather than to find a susceptible animal to which the so-called therapeutic dose would be toxic.

It is probable that if other drugs are fed in large quantities similar changes in the bone marrow would be produced. However, such a

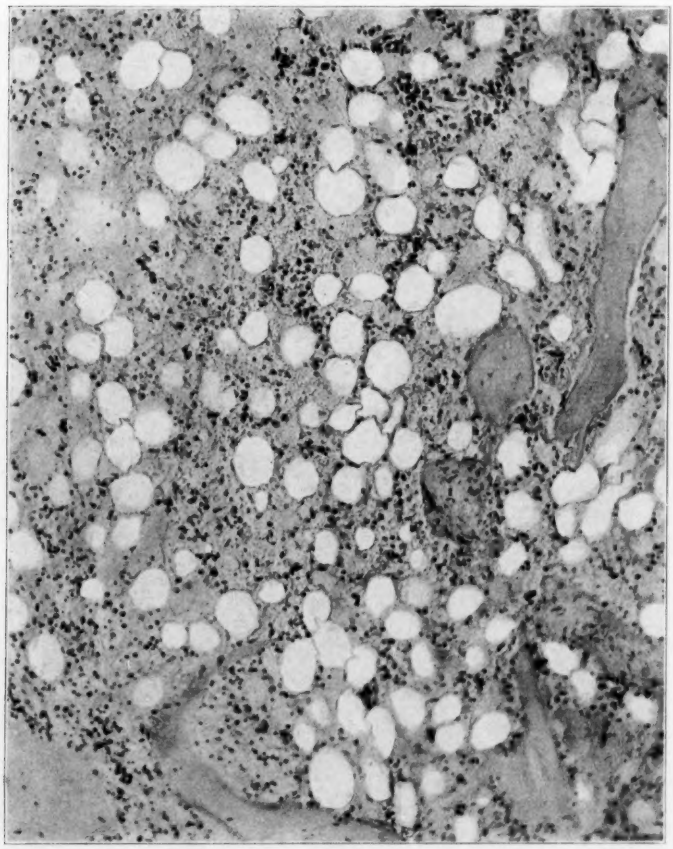


Fig. 6 (dog 7).—Bone marrow (× 160).

probability does not in any way detract from the fact that aminopyrine, aside from its direct systemic toxic action, has a selective action on bone marrow.

The transference of these experimental results from dogs to man would be extremely hazardous and unscientific had we no more informa-

tion than is found in our experiments. However, there is considerable evidence to show that a small percentage of persons are extremely sensitive to aminopyrine and react by the development of neutropenia that may result in death. These and other facts found in our summary

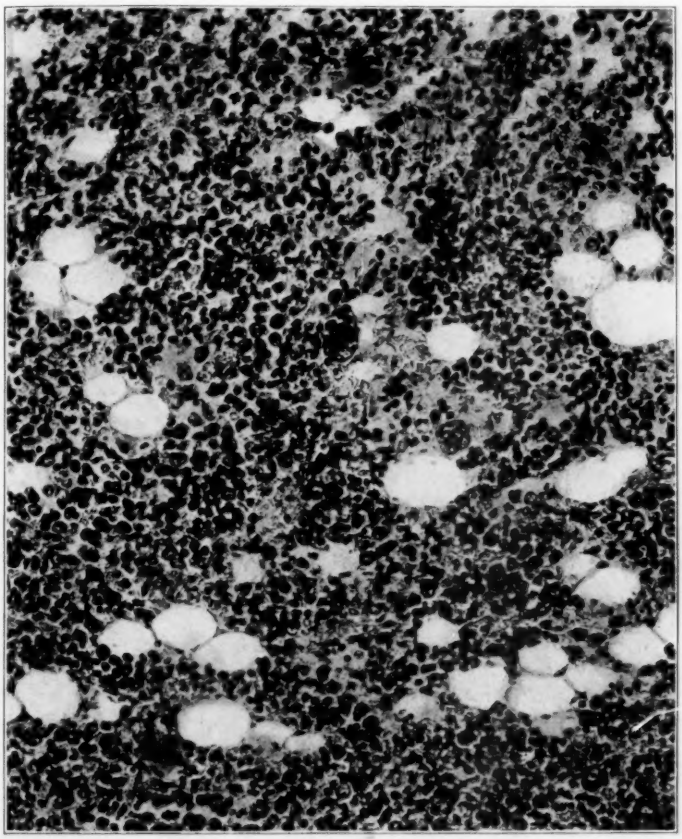


Fig. 7 (control dog).—Bone marrow of the tibia (× 310).

of the literature clearly establish aminopyrine as one of the causes of malignant neutropenia. Our experimental results merely establish the fact that aminopyrine has a selective toxic action on bone marrow.

It is our opinion that aminopyrine is only one of a variety of agents causing malignant neutropenia, although an important one.

CONCLUSIONS

It has been shown in these experiments that aminopyrine has a toxic effect on the bone marrow of dogs when administered orally in large doses.

The end result is one of severe aplasia of the bone marrow, rather than of hyperplasia with "maturation arrest."

Neutropenia was noted in 1 of the 2 dogs having a markedly aplastic bone marrow.

A summary of the literature on the experimental production of malignant neutropenia is presented.

ANATOMIC OBSERVATIONS ON SEVENTY HOSPITAL PATIENTS AFTER SUDDEN DEATH

JAMES R. LISA, M.D.
AND
JAMES FINLAY HART, M.D.
NEW YORK

Between Jan. 1, 1927, and June 30, 1936, autopsies were performed on 70 patients who died suddenly in the wards of the New York City Hospital. All of these patients were sufficiently sick to require hospitalization, some being dangerously ill while others were ambulatory and comfortable. Death was sudden in the respect that it came on unexpectedly and occurred within twenty minutes of the initial symptoms. We have analyzed and grouped these cases, and we hereby report our observations.

The great majority of the deaths were cardiac. There were 49 patients, over two thirds of the total number, who gave anatomic evidence of fatal changes in the heart. The histologic data on many of these patients have been reported in detail by one of us.[1] In 7 instances the cause of death was cerebral and in 4 pulmonary. Spontaneous rupture of the aorta accounted for 3 fatalities, ruptured aneurysm for 3, gastric accidents for 2, asphyxiation by a bolus of food for 1 and status lymphaticus for 1.

The cardiac causes could be arranged in eight subdivisions. Acute miliary infarction was the most prominent, being noted twelve times. Acute toxic myocarditis came next, with 10 instances. Acute coronary thrombosis followed, with 9 examples, and acute infectious myocarditis, with 7. The division of acute rheumatic infection consisted of 4 cases, acute interstitial involvement of 3, heart block of 2 and aortic stenosis of 1.

While we were able to arrange these cases in eight subdivisions, it was accomplished with no little difficulty. For example, 2 of the cases of acute miliary infarction could have been justifiably listed under infectious myocarditis. Bacterial emboli were noted in the myocardial branches of the coronary arteries in 1 case, and in another there was acute infectious endocarditis of coccic orgin with thrombi in the coronary arterioles. Again, of the cases of interstitial

From the First Medical Division, service of Dr. John Carroll, and the Pathological Laboratories, New York City Hospital, Department of Hospitals.

1. Lisa, J. R.: The Pathology of the Heart in Sudden Cardiac Death, Ann. Int. Med., to be published.

myocarditis, the condition in 1 was associated with acute lobar pneumonia, acute pericarditis and acute peritonitis. It would have been justifiable to include this with cases of infectious myocarditis. Furthermore, in 1 case listed under acute toxic myocarditis the disease was associated with acute lobar pneumonia, acute mediastinitis and acute pericarditis. The last 2 cases were not classed with those of infectious myocarditis, solely because organisms were not demonstrable within the myocardium.

The lesion we call acute miliary infarction has been reported by Roesler and Soloff.[2] It consists of a small area of myocardial necrosis about the size of a low power microscopic field, sharply delimited and consisting practically of preexisting reticulum only, with remnants of myocardial cytoplasm, nuclei and lipochrome. Occasionally a few degenerating polymorphonuclear cells are seen at the periphery.

In all of our cases of coronary thrombosis the patients were in the old age group and showed extreme arteriosclerosis of the coronary arteries. In 6 cases only had there been associated clinical evidence of hypertension and arteriolosclerosis of the kidneys. Two patients were Negroes, a man and a woman, although coronary thrombosis is commonly believed to be a rarity in this race.

We observed 9 cases of acute toxic myocarditis and 8 cases of acute infectious myocarditis. In 5 cases of the infectious group the condition was associated with acute valvular endocarditis. In 3 of the cases there was syphilitic myocardial involvement, in 1 of which there was an acute rheumatic myocarditic lesion almost as extensive as the syphilitic lesion. In another case an unusual syphilitic manifestation consisted of arteritis affecting the myocardial branches of the coronary arteries, with softening proceeding to local thrombosis. In 6 of the 9 cases of toxic myocarditis there was associated acute pneumonia; in 1 of these cases streptococci were isolated, and in 1 there was a history of a recent infection of the upper respiratory tract.

The 4 cases of acute rheumatic myocarditis were characterized by an extraordinary number and wide distribution of Aschoff bodies. While sudden death in acute rheumatic heart disease is uncommon, it has been previously reported.[3]

The patients with heart block exhibited the Stokes-Adams syndrome. In 1 case the condition was complicated by a meningioma of the dura, while in the other only very mild fatty myocardial degeneration was apparent. Although the heart block was present in the latter case for over a year, the conduction system failed to show any histologic changes.

2. Roesler, H., and Soloff, L. A.: Ann. Int. Med. **9**:477, 1935.

3. Rheumatic Heart Disease, Cabot Case 22041, New England J Med. **214**: 154, 1936.

In the case of aortic stenosis there was extremely tight stenosis of the aortic valve with minimal lesions deep in the myocardium accompanied by lymphoid, plasma and a few polymorphonuclear cells.

In 4 of the 7 cases of cerebral involvement the causes of death were proved at autopsy. They were, respectively, recent hemorrhage into the medulla, Torula meningoencephalitis, pontile hemorrhage and tumor of the brain. In the remaining 3 cases the general autopsy failed to reveal the cause of death, and the brains were not obtained; observation, however, suggested the probability of cerebral lesions.

The pulmonary group consisted of 4 cases. In 3 of these death was due to pulmonary embolism. The origin of the embolism was not determined in 2 cases, but in the third it was observed to arise from extension into the vena cava of primary carcinoma of the kidney.[4] In the last case there was a massive hemorrhage from pulmonary tuberculosis.

The ruptured aneurysms were all syphilitic. Two were thoracic and one was abdominal. Although this appears inconsistent with the known tendency for abdominal aneurysms to rupture, in the series of syphilitic aneurysms examined post mortem in the hospital, there was a much higher percentage of abdominal than of thoracic ruptures.

In 2 of the 3 cases of acute rupture of the aorta the rupture had occurred in the intrapericardial portion of the aorta and resulted in hemopericardium. In both there was associated essential hypertension. In 1 there was the complication of syphilis, but this had affected the aorta well beyond the scene of rupture. In the third case the abdominal aorta was involved, and the point of rupture was near an arteriosclerotic plaque. It was of interest to note that in 1 case of thoracic rupture an infectious myocardial lesion was demonstrable histologically and that in the case of abdominal rupture extensive septicopyemia was present.

In 1 case of gastric involvement death resulted from hemorrhage in a carcinoma; in another necropsy revealed perforation of a gastric ulcer with peritonitis.

Asphyxia caused by a bolus of food in a patient with bulbar palsy accounted for 1 death, while another was due to status lymphaticus in a pregnant woman who was being anesthetized.

RELATION OF POSTMORTEM TO CLINICAL DIAGNOSIS

We were interested to find in what percentage of cases the clinical diagnosis was correct. In 26, or about one third, of the cases the anatomic findings corroborated the clinical diagnosis. In 9 instances the clinical diagnosis was not correct, but could be considered reason-

4. Boyd, C. S., and Lisa, J. R.: J. Pediat. **5**:608, 1934.

able. In the remaining 35 cases the patients died of conditions not recognized by the clinicians.

In the group of conditions that were correctly diagnosed, the outstanding involvement was acute coronary thrombosis, which occurred in 8 cases. The clinical picture of this is distinctive, and as it is a commonly recognized cause of sudden death one would not be surprised to find a considerable degree of accuracy in the diagnosis. In the cases of acute rheumatic myocarditis the disease was diagnosed clinically, while apoplexy, heart block and ruptured syphilitic aneurysm were each recognized as the cause twice. In the remaining 9 cases the conditions varied in type and were easily recognized clinically.

In 3 cases in which we designate the clinical diagnosis as reasonable a condition diagnosed as coronary sclerosis was seen to consist of acute miliary infarctions of the heart. In 1 a condition diagnosed as spontaneous pneumothorax was a pulmonary embolism; in 1 a possible abscess of the lung was actually a tuberculous hemorrhage, and in 1, acute rheumatic fever, with presenting migrating arthritis for three weeks, proved to be acute endocarditis and myocarditis, probably meningococcic. In 1 case a condition diagnosed as tuberculous meningitis was Torula meningoencephalitis, and in a case in which the diagnosis was toxic goiter with coronary sclerosis an inflammatory lesion of the myocardium was present. A patient said to have pernicious anemia who died of a cardiac condition was considered at autopsy to have had acute toxic myocarditis.

The clinical diagnosis in the remaining 35 cases did not explain the cause of death. In 25 cases the anatomic diagnosis was myocarditis. Of these, instances of miliary infarctions comprised the largest group, with toxic myocarditis second and infectious myocarditis third.

The clinical features of acute miliary infarctions are those of acute left ventricular failure. Miliary infarctions are usually diagnosed as coronary thrombosis because they occur most frequently in cases of recognized coronary sclerosis, and at autopsy they are practically always associated with the more severe grades of coronary sclerosis. Acute toxic myocarditis is usually associated with pneumonia. This is not generally appreciated by clinicians, although Gonzales found it occasionally during the pandemic of 1918-1919.[5] Infectious myocarditides are not diagnosed because other factors, such as syphilis and rheumatic heart disease, are more prominent and mask the features of the infectious element. In our experience at the hospital infection superimposed on valves deformed by syphilis or rheumatism is much more frequent than the literature indicates. On clinical grounds, this con-

5. Gonzales, T. A.: Personal communication to the authors.

dition can frequently be suspected because a sudden increase in cardiac symptoms occurs after a long period of slight variability.

MAIN CAUSE OF DEATH A VASCULAR CONDITION

Our observations lead us to believe that the principal cause of sudden death may be considered as vascular. This is especially true when we employ "vascular" in the broad sense and include the heart as a modified blood vessel. Of the entire series, in 62 cases involvement was definitely of this character. In 5 cases only the condition was nonvascular, with 1 instance each of asphyxiation from a bolus of food, Torula meningoencephalitis, brain tumor, perforated gastric ulcer and status thymicolymphaticus. In the remaining 3 the cause probably was a cerebral vascular accident of embolic nature.

ROLE OF INFECTION

It was apparent that infection played a determining part in many of these cases of sudden death. In the majority of cases in the cardiac group there was evidence of an infectious or toxic factor affecting the myocardium. In 12 cases of the infectious group syphilis, rheumatism and the acute endocarditides accounted for death, in all with profound damage to the myocardium. The most prominent feature of the cases in the toxic group was the presence of acute pulmonary infections, which occurred in the great majority. This factor was particularly noticeable in infants and children. In the older patients the good condition of the coronary vessels was striking. Acute or chronic pulmonary infections were prominent also in the cases in which there was acute interstitial myocarditis. It was of interest to note that in 1 instance each in the infectious and in the toxic group acute pericarditis complicated the pneumonia. In a somewhat smaller percentage, but in more than half, of the cases of miliary infarcts, chronic pulmonary infections were present. In 1 instance bacterial masses were seen in the myocardial arterioles of the walls affected. In a second case acute arteriolar thrombi were present with acute endocarditis. We noted in the majority of the cases of this group the most severe grade of sclerotic changes of the coronary arteries. A toxic element was more difficult to prove in the cases of acute coronary thrombosis, but in 2 instances an acute glomerular lesion suggested that such a factor was present.

Spontaneous rupture of the aorta is generally believed to be due to acute medial necrosis of toxic origin, which occurs in an otherwise normal vessel. In 1 example in the present series, clinically mild pharyngitis had preceded by a few days the vascular accident. In this case an odd histologic finding in the heart was acute infectious myo-

carditis. In 2 cases of miliary infarction, in both with minimal coronary arteriosclerosis, acute medial necrosis of the coronary vessels was associated with purulent bronchiectasis. This lesion is similar to that which results in spontaneous rupture of the aorta.

In the case of rupture of the arteriosclerotic aorta, a bacterial cause was not demonstrated. The septicopyemia, however, strongly suggested that a local septic process had been present and had been destroyed by subsequent massive hemorrhage.

SUMMARY AND CONCLUSIONS

Considering this report from a clinician's point of view, we note that about one third of the deaths were explainable by the clinical diagnosis. There were also 9 cases in which the diagnosis was nearly enough correct to be considered reasonable. One may say that the chances of making a correct diagnosis of sudden death under similar conditions are about 1:2. It is possible that the percentage of errors might be improved, as some of the patients whose condition was incorrectly diagnosed were not long enough in the hospital for a thorough study to be made.

As one would expect, we found sudden death to be largely the result of a vascular accident. In about 89 per cent of our cases the cause was proved to be of this nature. Infection played a recognizable role in these fatalities. Aside from syphilis, rheumatism and acute endocarditis, we observed that acute or chronic pulmonary infections were commonly associated. This was particularly noticeable in children.

We believe that one might arrive at a more perfect diagnostic score by giving greater consideration to the possibilities of the vascular, or perhaps the cardiovascular, aspect, especially when infection is present. In 25 of the 35 cases in which the cause was unrecognized death was attributed to myocarditis. In most if not all of these there was clinical evidence of concurrent infection. Had the clinician been more conscious of the possibility of acute miliary infarction of the myocardium and acute toxic myocarditis in cases of this type, there would have been a larger percentage of correct diagnoses. It would be reasonable to say that in 55 or more of the 70 cases the condition could have been correctly diagnosed if these pathologic changes were more commonly known to clinicians.

ROENTGENOGRAMS OF THE CHEST AND THE INTRACUTANEOUS TUBERCULIN TEST FOR ADULTS

A COMPARATIVE STUDY

LUKE W. HUNT, M.D.
CHICAGO

For many years investigators have striven to evolve some sort of test which would help in the diagnosis of tuberculosis. In 1907 von Pirquet brought forth his well known cutaneous test, and soon several modifications of the test were introduced, including the Mantoux intracutaneous test, which has so well stood the test of time that at present it is used more extensively than any of the others. The value of the Mantoux test as an aid in establishing a diagnosis of tuberculosis in infancy and early childhood has been accepted for many years. Reports of its value in adult life have been received with varying degrees of approbation and distrust, and only in recent years has it been found to have a definite place, when properly performed and interpreted, as an aid in establishing a diagnosis of tuberculosis in older patients.

The lack of agreement as to the value of the intracutaneous tuberculin test for adults may be due to the facts that it is generally used as a qualitative test and that in routine testing dilutions of 1 : 100, 1 : 1,000 and 1 : 10,000 are used. There is, moreover, no general agreement as to what constitutes a positive reaction. Atsatt [1] in reporting a series of tuberculin tests on 211 patients with tuberculosis of bones and joints stated that the reactions were read in 6 degrees of intensity but did not describe the reaction which he interpreted as positive. He used dilutions of 1 : 1,000, 1 : 2,500, 1 : 5,000 and 1 : 10,000; the 1 : 10,000 dilution was considered as diagnostic. King,[2] in another series of tests, using a 1 : 20,000 dilution as diagnostic, considered a positive reaction to be an area of erythema, nearly always indurated, 1 cm. or more in diameter. Blair and Galland [3] stated that their criterion for a positive reaction

From the Department of Medicine, University of Chicago.

1. Atsatt, R. F.: Studies with a Quantitative Tuberculin Reaction, J. Bone & Joint Surg. **9**:657 (Oct.) 1927.

2. King, R. B.: A Tuberculin Test of Value in Adults, New England J. Med. **207**:831 (Nov. 10) 1932.

3. Blair, J. E., and Galland, W. L.: A Differential Quantitative Tuberculin Test, Am. Rev. Tuberc. **23**:1 (Jan.) 1931.

is the formation of an indurated cutaneous nodule, whatever its size, usually accompanied by erythema. They used dilutions of 1 : 10,000, 1 : 100,000, 1 : 1,000,000 and 1 : 10,000,000. The 1 : 100,000 dilution was considered diagnostic. Ayman,[4] using dilutions of 1 : 10,000 to 1 : 10,000,000, with the 1 : 50,000 dilution as diagnostic, considered a positive reaction to be an area of erythema 10 by 9 mm. or more, nearly always indurated. Moncrieff,[5] reporting a series of qualitative tuberculin tests of children with dilutions of 1 : 100, 1 : 1,000 and 1 : 10,000, stated that a positive reaction consists of a red area of swelling, easily palpable, frequently tender and with an average diameter of 2 cm. He also stated that erythema alone without a palpable swelling is not sufficient for a reaction to be called faintly positive. Opie and McPhedran [6] in their investigation assumed that an area of redness 10 mm. or more in diameter indicates a positive reaction and stated that "inconspicuous spots of redness or redness with no edema have not been regarded as definite evidence of hypersensitiveness to tuberculin." Hart,[7] on the other hand, stated that "a reaction should be regarded as positive if it consists of an area of erythema or erythematous infiltration the greatest diameter of which equals or exceeds 5 mm." Cummins and Walker [8] even considered that "no reaction measuring less than 2 mm. across can be accepted as definite."

Not only is there no general agreement as to what constitutes a positive tuberculin reaction, but some of the technics reported in the literature are open to question. In some instances the tests have been performed by different persons and the results interpreted by nurses and interns; unless such workers have had a good deal of experience with the tests the percentage of error is likely to be high. Cummins and Walker stated that their object was to raise a wheal about the size of a threepenny piece, and they paid more attention to the attainment of this effect than to the absolute amount of fluid injected, although the average volume was "round about 0.1 cc."

It is generally agreed that the qualitative tuberculin test with the dilutions commonly used is of value in establishing a diagnosis of

---

4. Ayman, D.: The Intracutaneous Quantitative Tuberculin Test in the Diagnosis of Active Tuberculosis, J. A. M. A. **103**:154 (July 21) 1934.

5. Moncrieff, A.: The Clinical Interpretation of the Intradermal Tuberculin Reaction, Quart. J. Med. **24**:153 (Jan.) 1931.

6. Opie, E. L., and McPhedran, F. M.: The Contagion of Tuberculosis, Am. Rev. Tuberc. **14**:347 (Oct.) 1926.

7. Hart, P. D. A.: The Value of Tuberculin Tests in Man, Medical Research Council, Special Report Series, no. 164, London, His Majesty's Stationery Office, 1932, p. 126.

8. Cummins, S. L., and Walker, S. B. H.: Investigation and Assessment of One Hundred Cases of Pulmonary Tuberculosis, Tubercle **12**:436 (July) 1931.

tuberculosis only for infants and very young children and is of extremely limited value for adults. The quantitative factor, or the measure of the degree of hypersensitivity to tuberculin, has been greatly neglected. From the time of the earliest description of the tuberculin reaction there has been a semblance of the belief that there is a relation between the degree of sensitivity to tuberculin, as shown by the cutaneous reaction, and the extent of the tuberculosis in the person being tested. Thus von Pirquet required for a positive reaction an area of erythema not less than 5 mm. in diameter. Mantoux's technic demanded the intradermal injection of 1 drop of a 1:5,000 dilution of tuberculin. Both investigators thus presupposed a quantitative element in these tests. It appears plausible that active tuberculous disease, with continual reaction of the patient to repeated liberation of toxic material from the focus of activity, is accompanied by a degree of hypersensitivity different from that associated with a latent infection or a long-continued process of healing, in which the escape of toxic substance from a focus is inhibited or prevented by walling off. By administration of a series of graded dilutions of tuberculin it should be possible to establish a point above which reactions can be obtained only in those who possess a considerable degree of hypersensitivity, that is, persons actively combating tuberculous disease.

This paper describes an effort to correlate the local reactions to the intracutaneous injections of different dilutions of commercial old tuberculin with the roentgen findings in the chests of 460 healthy young adults and 45 young adults with active lesions, the object being to evaluate the cutaneous reactions in terms of past or present tuberculous infection in the lungs. It also includes a brief discussion of some of the problems encountered in such a study.

MATERIAL

Two groups of young adults were used for the study. The first group consisted of 232 nurses ranging in age from 18 to 30 years. They were in good health at the time of the tests and had passed physical examinations when they began training. They were considered a homogeneous group, perhaps representing better health than a group of the same age chosen at random. The second group consisted of 230 healthy graduate nurses, interns and attendants and 43 adults who were found to have active lesions on routine physical examination. Their ages varied from 20 to 35 years; approximately 95 per cent were between 20 and 30.

METHOD

The tuberculin tests were performed by the intradermal injection of 0.1 cc. of different dilutions of commercial old tuberculin into the flexor surfaces of the forearms and one injection of 0.1 cc. of the diluting fluid for control. The diluting fluid contained 0.2 per cent phenol in an 0.85 per cent solution of sodium chloride. For the first group 1:1,000 and 1:5,000 dilutions were used and for the second

group 1:10,000 and 1:100,000. The dilutions were made fresh each week. MacGregor syringes containing 1 cc. and graduated in tenths and 27 gage needles were used. All reactions to tests and controls were measured for induration and redness after twenty-four and forty-eight hours and the majority also after seventy-two hours. All the tests and readings for the two groups were done by the same person.

The reactions were divided according to severity into the following groups:

Group	Reaction (Induration), Mm.
0	None
1	10 by 10 to 15 by 15
2	15 by 15 to 20 by 20
3	20 by 20 to 30 by 30
4	30 by 30 and above

The roentgen plates of the chests were studied with regard to signs of past and present infection. Naturally in this group many of the plates were considered normal from the roentgenologic standpoint, but the following divisions were made:

Group	Reading
I	Completely negative
II	Large irregular calcifications in hilus, parenchyma or both
III	Signs of present activity

The results are recorded in the table. It will be seen that in group I, those whose roentgenograms were completely negative, 131, or 62.4 per cent, gave a negative reaction to the 1:1,000 dilution and 79, or 37.6 per cent, a positive reaction; with the 1:5,000 dilution, 169, or 80.4 per cent, gave a negative and 41, or 19.6 per cent, a positive reaction; with the 1:10,000 dilution, 176, or 83.8 per cent, gave a negative and 34, or 16.2 per cent, a positive reaction; with the 1:100,000 dilution, 203, or 96.7 per cent, reacted negatively and 7, or 3.3 per cent, positively. In group II, those with large irregular calcifications in the hilus or parenchyma, 9, or 45 per cent, gave a negative reaction and 11, or 55 per cent, a positive reaction to the 1:1,000 dilution. With the 1:5,000 dilution, 12, or 60 per cent, had a negative and 8, or 40 per cent, a positive reaction; with the 1:10,000 dilution, 15, or 71.5 per cent, had a negative and 5, or 28.5 per cent, a positive reaction and with the 1:100,000 dilution, 20 had a negative and none a positive reaction.

In group III, those with roentgen evidence of present activity, only 2 were tested with the 1:1,000 and the 1:5,000 dilution, and both had positive reactions. With the 1:10,000 dilution and the 1:100,000 dilution, 41, or 94.8 per cent, gave a positive and 2, or 4.6 per cent, a negative reaction. Hence with 1:1,000 and 1:5,000 dilutions of old tuberculin there seems to be no correlation between the cutaneous reactions and the roentgen findings of the chests of young adults sufficient to

Comparison of Roentgenograms of the Chest and Intracutaneous Tuberculin Reactions of Five Hundred and Five Young Adults with 1: 1,000, 1: 5,000, 1: 10,000 and 1: 100,000 Dilutions of Old Tuberculin

Group	Roentgen Findings	1:1,000 Dilution						1:5,000 Dilution					
			Reaction (Induration), Mm.						Reaction (Induration), Mm.				
		Number of Negative Results	10 by 10 to 15 by 15	15 by 15 to 20 by 20	20 by 20 to 30 by 30	30 by 30 or Above	Total	Number of Negative Results	10 by 10 to 15 by 15	15 by 15 to 20 by 20	20 by 20 to 30 by 30	30 by 30 or Above	Total
I	Completely negative.....	131 (62.4%)	16	13	13 (37.6% positive)	43	210	169 (80.4%)	19	13	9 (19.6% positive)	0	210
II	Large irregular calcifications in hilus, parenchyma or both	9 (45%)	0	1	2 (55% positive)	8	20	12 (60%)	5	3	0 (40% positive)	0	20
							230						230
III	Signs of present activity...	.....	..	1	1	..	2	...	0	1	1	0	2
		1:10,000 Dilution						1:100,000 Dilution					
I	Completely negative.....	176 (83.8%)	23	11	0 (16.2% positive)	0	210	203 (96.7%)	3	4	0 (3.3% positive)	0	210
II	Large irregular calcifications in hilus, parenchyma or both	15 (71.5%)	4	1	0 (28.5% positive)	0	20	20 (100%)	0	0	0	0	20
							230						230
III	Signs of present activity...	2 (5.2%)	0	11	14 (94.8% positive)	16	43	2 (5.2%)	7	13	12 (94.8% positive)	9	43

be of worth in evaluating the past or present activity of tuberculous infection in the lungs. But response to a dilution of 1 : 100,000 may usually be considered indicative of active tuberculosis, and response to the 1 :10,000 and not to the 1 : 100,000 dilution seems to indicate latent or healed tuberculosis.

It should be stated that of the 7 subjects who had negative roentgenograms and who had a positive reaction to the 1 : 100,000 dilution, 3 had a history of contact: One, a nurse, had just finished a course of training at a tuberculosis sanatorium; another, an intern, had finished an internship at a similar institution, and the third was a man whose wife had tuberculosis. One of the others had rather extensive calcifications in the hilus and parenchyma. The significance of definite evidence of calcification in the hilar region or in the parenchyma has only recently been appreciated. Pathologists have shown that unless these lesions are completely calcified they should not be looked on as healed. For a discussion of this subject the reports of Hektoen,[9] Opie and Aronson[10] and, more recently published, Feldman and Baggenstoss[11] should be consulted. At any rate, the roentgenogram gives little information on what is happening within and immediately around these deposits. This is well illustrated by 1 of the group, who had a positive reaction to the 1 : 100,000 dilution and whose roentgenograms showed extensive calcification in the hilus and parenchyma. The roentgenograms were considered to show normal conditions except for the calcifications. About a year after the first plates were taken an exploratory laparotomy was done, and the patient was found to have tuberculous peritonitis. Roentgenograms made on several occasions before the operation showed no change from the original plates, but the primary focus was probably in the lungs.

The 2 adults who had roentgen evidence of active lesions and whose tuberculin reactions were negative had small areas of infiltration about 3 mm. in diameter in the subapical region but no other evidence of tuberculosis. Unfortunately, further testing of these subjects could not be done.

It should be stated also that the majority of the patients with active lesions had a minimal exudative type of tuberculosis. Westwater[12]

9. Hektoen, L.: Tubercle and Morbid Anatomy, in Klebs, A. C.: Tuberculosis: A Treatise by American Authors, New York, D. Appleton and Company, 1909, chap. 2, pp. 52-79.

10. Opie, E. L., and Aronson, J. D.: Tubercle Bacilli in Latent Tuberculous Lesions and in Lung Tissue Without Tuberculous Lesions, Arch. Path. **4**:1 (July) 1927.

11. Feldman, W. H., and Baggenstoss, A. H.: The Residual Infectivity of the Primary Complex of Tuberculosis, Am. J. Path. **14**:473 (July) 1938.

12. Westwater, J. S.: Allergy in Tuberculosis: The Quantitative Use of the Mantoux Test, Tubercle **15**:543 (Sept.) 1934.

concluded from his work that the young adult with exudative disease tends to have higher sensitivity than the older subject with the fibroid type, i. e., the more recent and active the disease, provided it is not massive, the higher the sensitivity.

In the group with active lesions the cutaneous reactions were usually larger than in the other groups. None of the positive reactions in the group with active lesions measured less than 10 by 10 mm. However, the size of the reaction is perhaps not as important as the degree. It is obvious that an area of 20 mm. in diameter showing simple infiltration means less sensitivity than a reaction of the same size showing necrosis. In our experience induration is the most important criterion of a positive reaction. Erythema alone is unreliable. It is occasionally seen in sensitive skins; in case of Negro patients it is often impossible to judge the presence of erythema. In this series, at least, the most intense reactions showed induration, pale yellow edema and occasionally vesiculation.

These results indicate that the quantitative tuberculin test is a valuable confirmatory diagnostic procedure in early adult life and is perhaps as valuable an aid as the qualitative test in infancy and early childhood. In case of infants and young children one is usually dealing with the first infection type of tuberculosis, while in case of adults it is the reinfection type. To use the words of von Pirquet, the first group as a rule has not "met the tubercle bacillus," while the second group has done so. It is well known that certain conditions modify the tuberculin reaction, but these conditions are usually obvious and have to be taken into consideration when one interprets the test. It is also well known that a tuberculin test and a roentgen examination of the chest do not constitute complete examination for tuberculosis: They only provide evidence that may be used by the clinician in conjunction with the history and with results of physical and laboratory examination in arriving at a final diagnosis. However, probably the best method of selecting subjects for such an examination is the use of the quantitative tuberculin test and roentgenograms.

CONCLUSIONS

Intracutaneous tuberculin tests were performed on a group of 505 young adults with different dilutions of old tuberculin, and the results were compared with roentgen evidence of past or present tuberculous infection in the lungs.

With 1:1,000, 1:5,000 and 1:10,000 dilutions of old tuberculin there was not sufficient correlation between the cutaneous reactions and the roentgen findings to be of worth in evaluating the past or present activity of tuberculous infection in the lungs.

With the 1 : 100,000 dilution, however, 96.7 per cent of 230 young adults whose roentgenograms were negative gave a negative reaction and 3.3 per cent a positive reaction. With the same dilution, 41, or 94.8 per cent, of 43 young adults who had roentgen and other evidence of active tuberculosis gave a positive reaction and 2, or 5.2 per cent, gave a negative reaction.

None of the positive reactions in those with active lesions had an area of induration measuring less than 10 by 10 mm. Induration is considered the most important criterion of a positive reaction. Erythema alone is unreliable.

The quantitative intracutaneous tuberculin test is a valuable aid when used in conjunction with other available data in establishing a diagnosis of tuberculosis in young adults. Its accuracy when it is properly performed and interpreted is comparable to that of the commonly accepted immunologic and serologic diagnostic procedures.

TEST OF GLOMERULAR FUNCTION WITH SODIUM FERROCYANIDE

FURTHER STUDIES

EDWARD J. STIEGLITZ, M.D.
GARRETT PARK, MD.

The first clinical studies on the urinary excretion of sodium ferrocyanide as a criterion of glomerular efficiency were reported in 1934.[1] It was then shown that the curves for the excretion of this salt by normal persons are characteristic and have but little variation. In the succeeding four years additional data have been obtained, and these continued studies have further clarified the application of the test.

METHOD

There has been no significant change in the technic proviously reported. Ampules of sterile dry pure sodium ferrocyanide containing 0.5 Gm. of the hydrated salt, or about 0.25 Gm. of the anhydrous salt, were employed. Ferrocyanide salts might be decomposed by the gastric acid if taken orally, but on slow intravenous injection no evidences of toxic effects have been observed.

The contents of one ampule are dissolved in 10 cc. of sterile distilled water and administered by slow intravenous injection. It is essential that this solution be clear before the injection is started; the salt dissolves rather slowly. Specimens of urine are collected thirty, sixty, one hundred and twenty and one hundred and eighty minutes after the injection and their content of ferrocyanide determined by analysis. It is immaterial just how much water is consumed before or during the test; the rate of excretion appears to be independent of the volume of urine. Ample fluid to make possible the prompt voiding of urine at the proper intervals is desirable.

The titration of ferrocyanide is carried out with a 0.4 per cent solution of cupric sulfate (0.004 Gm. per cubic centimeter). Before titration the specimen is acidified with concentrated sulfuric acid. Cupric ferrocyanide is a slightly soluble red salt but is more soluble than the well known prussian blue (ferriferrocyanide). To determine the end point in titration, drops of solution of ferric chloride are placed on a tile, and as the titration proceeds a drop of the unknown is placed in contact with the ferric chloride. If free ferrocyanide is present prussian blue is imme-

---

From the Department of Medicine of the Chicago Memorial Hospital.

Ampules of sterile dry pure sodium ferrocyanide were supplied by the Abbott Laboratories.

1. Stieglitz, E. J., and Knight, A. A.: Sodium Ferrocyanide as Clinical Test of Glomerular Efficiency: Preliminary Report, J. A. M. A. **103**:1760 (Dec. 8) 1934.

diately formed; if all the ferrocyanide has already been precipitated as cupric ferrocyanide, a distinct delay occurs before the appearance of this blue color. The end point is therefore read when the appearance of the blue is delayed four to five seconds.

The calculation is based on the chemical equation: $Na_4Fe(CN)_6 + 2\ CuSO_4 \rightarrow Cu_2Fe(CN)_6\downarrow + 2\ Na_2SO_4$. The molecular weight of $Na_4Fe(CN)_6$ is 304.08 and that of $CuSo_4 \times 2$ is 319.26. If the solution of cupric sulfate contains *exactly* 0.004 Gm. per cubic centimeter, then 1 cc. of the solution precipitates exactly 0.0038 Gm., or 1.52 per cent, of the injected dose of 0.25 Gm. of anhydrous sodium ferrocyanide. The exact strength of the cupric sulfate is best first measured by electrolytic methods.

REVIEW OF PREVIOUS WORK

The application of sodium ferrocyanide to clinical study of the glomerular efficiency of the kidneys is based on the premise that ferrocyanide salts are excreted solely by the glomeruli. This almost unique distinction was first noted by Marshall and Grafflin [2] when they observed that the practically aglomerular kidneys of Lophius piscatorius eliminated no ferrocyanide or only 0.2 to 0.5 per cent of the quantity eliminated by fish with glomerular kidneys.[3] Histochemical studies on mammalian kidneys by Gersh and Stieglitz,[4] who utilized the highly accurate Altmann freezing-drying technic,[5] or rapid perfusion with a solution of cupric sulfate to fix the ferrocyanide, revealed that in the normal rabbit ferrocyanide is eliminated entirely via the glomeruli. No evidence of tubular excretion was discovered. Identical distribution of ferrocyanide in the tissues of the kidneys of dogs and monkeys has since been observed by Gersh.[6] The excretion of ferrocyanide ceases when arterial tension is reduced by section of the medullary portion of the spinal cord. This behavior is in sharp contrast to that of ferric salts, which are excreted by the proximal convoluted tubules and whose excretion continues despite acute hypotension,[7] but at a rate which corresponds to the characteristically great drop in excretion of creatinine which was observed on lowering the arterial tension in normal men by spinal anesthesia.[8]

Extensive studies by Van Slyke, Hiller and Miller [9] on the ferrocyanide clearance in dogs showed that ferrocyanide, inulin and creat-

2. Marshall, E. K., Jr., and Grafflin, A. L.: Bull. Johns Hopkins Hosp. **43**: 205 (Oct.) 1928.

3. Marshall, E. K., Jr.: Am. J. Physiol. **94**:1 (July) 1930.

4. Gersh, I., and Stieglitz, E. J.: Anat. Rec. **58**:349 (March 25) 1934.

5. Gersh, I.: Anat. Rec. **53**:309 (Aug. 25) 1932.

6. Gersh, cited by Van Slyke, Hiller and Miller,[9a] p. 613.

7. Stieglitz, E. J.: Am. J. Anat. **29**:33 (May) 1921. Holton, S. G., and Bensley, R. R.: ibid. **47**:241 (March) 1931.

8. Lassen, H. C. A., and Husfeldt, E.: J. Clin. Investigation **13**:263 (March) 1934.

9. Van Slyke, D. D.; Hiller, A., and Miller, B. F.: (*a*) Am. J. Physiol. **113**:611 and (*b*) 629 (Nov.) 1935.

inine follow the same excretory behavior, as their plasma clearances are equal and independent of the concentrations of plasma. They concluded that the plasma clearance of ferrocyanide, like that of inulin and/or creatinine, is equal to the glomerular filtration in dogs. Ferrocyanide and inulin were found not to enter the erythroplastids to any appreciable degree but to remain in the blood plasma, whereas urea diffuses rapidly to maintain intracellular equilibrium with the concentration of plasma.[9b] The urea clearance in dogs is about half the clearance of ferrocyanide, inulin or creatinine. It is assumed that urea is reabsorbed by the tubules whereas the other substances are not. Actually there is no convincing proof of the reabsorption of the solute by the tubules. That water (the solvent) is reabsorbed in the distal convoluted tubules is unquestioned. The assumption of the reabsorption of the solute by these tubules becomes necessary only when one utterly denies the existence of possible excretory activity on the part of the proximal convoluted tubules. Such denial is unwarranted.

Miller and Winkler [10] studied the sodium ferrocyanide clearance in man, using the analytic methods of Edwards and Langley.[11] Their results indicated close parallelism between the ferrocyanide clearances and the urea clearances, whereas the inulin clearances were nearly twice as great. These results are markedly different from those obtained with dogs,[9a] in which essentially identical excretory behavior was noted with creatinine, inulin and ferrocyanide. Miller and Winkler concluded that the mechanisms for excreting ferrocyanide in man and in the dog are not the same, that in man the salt may be reabsorbed (as urea is presumed to be) and that therefore the excretion of ferrocyanide in man does not necessarily parallel glomerular filtration. This apparent discrepancy requires confirmation of the data by different methods. It does not invalidate the clinical application of the excretion of ferrocyanide to the study of glomerular efficiency, for there is no evidence whatever indicating that ferrocyanide is excreted via any other route than through the glomeruli. In these studies with patients Miller and Winkler [10] employed doses of sodium ferrocyanide from three to twenty-four times larger than those originally used and recommended by Stieglitz and Knight.[1] These huge doses caused renal irritation.

Clinical comparison by Gordon [12] of the results of the test for excretion of sodium ferrocyanide with the results of a simultaneous urea clearance test revealed a coefficient of correlation of 0.7. It was observed also that the excretion of ferrocyanide did not alter the urea clearance. Gordon concluded that for practical clinical purposes the two tests gave

10. Miller, B. F., and Winkler, A.: J. Clin. Investigation **15**:489 (Sept.) 1936.
11. Edwards, J. G., and Langley, W. D.: J. Biol. Chem. **112**:469 (Jan.) 1936.
12. Gordon, W.: Am. J. M. Sc. **192**:208 (Aug.) 1936.

fundamentally parallel results. Baker and Habein,[13] employing the technic of Stieglitz and Knight, studied the excretion of ferrocyanide in cases of urinary obstruction. The reduction in excretion of ferrocyanide in their cases of prostatic obstruction paralleled the reduction of phenolsulfonphthalein output. Despite the severe renal impairment of many of their patients, no systemic toxic symptoms were noted in any case. It is remarkable, however, that when prostatic irritation exists, injection of ferrocyanide causes marked urethral and prostatic burning for one to two hours; the intensity of this local pain varies with the frequency of previous catheterization. The greater the urethral irritation and inflammation prior to the administration of ferrocyanide, the greater the discomfort. I also have observed this form of local reaction in elderly men and have refrained from employing sodium ferrocyanide for patients with known urethral irritation.

RESULTS

The previously reported series of tests made on persons with apparently normal renal function has now been extended to include tests on 150 such persons. The second series of tests yielded means slightly higher than those reported in 1934. The comparison of the results is shown in table 1.

A revised curve of the normal rate of excretion (chart 1) shows but minor shift from the rate previously considered normal. For the sake of convenience and clinical simplicity, it is adequate to consider the average normal output to be in the neighborhood of 15 per cent of the injected dose for the first thirty minutes, 25 per cent at the end of an hour and 35 and 45 per cent respectively after two and three hours.

Calculations based on the percentage of the injected dose excreted do not take into consideration the fact that the ferrocyanide available in the blood is constantly diminishing. There is no way of determining the rate of excretion in reference to the concentration of the blood except by "clearance" studies including determination of the ferrocyanide content of the blood.[9] In man this is not feasible, as the present methods for quantitation of ferrocyanide are too insensitive to permit measurement of minute quantities and therefore inadvisably large doses of the salt would be required. However, it is possible, with the present data, to correct the percentages of ferrocyanide excreted, allowing for that eliminated in the previous interval. It is true that this does not take into consideration the salt which may diffuse into tissues other than the kidneys. However, such diffusion as does occur probably takes place almost immediately after the injection of the salt and is therefore

13. Baker, G. S., and Habein, H. C.: Proc. Staff Meet., Mayo Clin. **11**:134 (Feb. 26) 1936.

a relatively constant source of error. When the calculations are revised the data unquestionably more closely approximate the true excretory rate than when the previously excreted ferrocyanide is ignored.

For example, in the normal person, 15.4 per cent of the injected ferrocyanide is eliminated in the first thirty minutes after injection and an additional 12.3 per cent in the second half hour. However, in this second interval only 84.6 per cent of the dose originally injected is theoretically available for excretion, and therefore the rate of excretion is actually higher than 12.3 per cent. For convenience and clarity, one may speak of the revised calculations as "corrected percentages."

TABLE 1.—*Ferrocyanide Output of Normal Patients* *

Time (Min.)	50 Patients (1934)		150 Patients (1938)	
30	13.5		15.4	
60	8.5	22.0	12.3	27.7
120	9.0	31.0	9.8	37.5
180	10.0	41.0	6.5	44.0

* Recorded in percentage of the injected dose excreted.

TABLE 2.—*Comparison of "Uncorrected" and "Corrected" Rates of Excretion of Normal Persons*

Time (Min.)	"Uncorrected" Normal Rate of Excretion (Percentage of the Injected Dose)	"Corrected" Normal Rate of Excretion (Percentage of the Salt Remaining in the Body)
30	15.4	15.4
60	12.3	14.54
120	9.8	13.55
180	6.5	10.41

Recalculation along such lines reveals some most suggestive data. A comparison of the uncorrected and the corrected percentages of excretion in shown in table 2.

As is to be expected, the decline in percentage of excretion of the theoretically available sodium ferrocyanide is much smaller. This relation is better visualized when these data are diagramed, as in chart 2. As previously stated, the "correction" does not take into consideration the diminution in the supply of ferrocyanide due to diffusion into tissues other than the kidneys, but this is probably not as serious a source of error as one first suspects. Ferrocyanide salts are extremely diffusible, and such disappearance from the blood stream into tissues probably takes place with great rapidity almost immediately after the intravenous injection of the salt. It is highly probable that after the first few minutes there exists a fairly stable equilibrium between the content of

ferrocyanide in the blood and that in the tissues. By far the greatest quantity of ferrocyanide is eliminated in the first hour.

Application of the test of glomerular function to the study of renal impairment in hypertensive arterial disease confirmed the findings

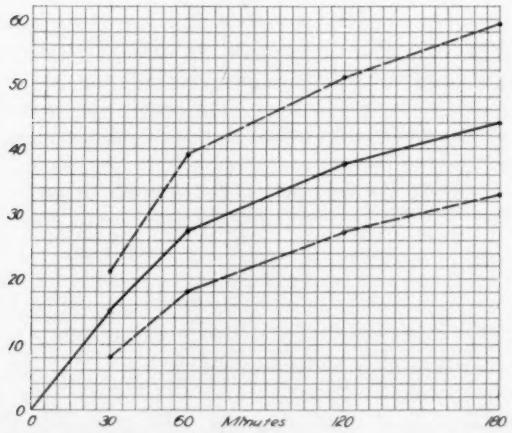


Chart 1.—Mean excretion of sodium ferrocyanide of 150 patients with presumably normal kidneys. The spread of variation is shown by dash lines. The values are recorded in terms of the percentage of the injected dose eliminated.

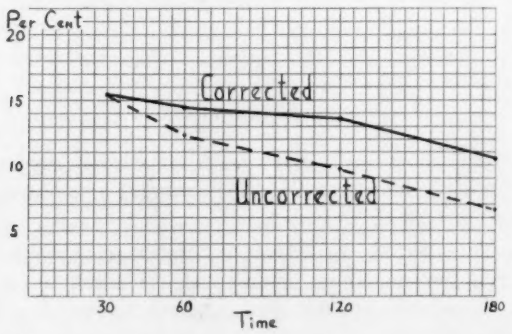


Chart 2.—Mean normal rates of excretion of ferrocyanide calculated in terms of percentage of the total injected dose (broken line) and "corrected" on the basis of the amount of ferrocyanide remaining within the body (solid line).

recorded in the preliminary report.[1] For some 80 additional cases of hypertensive disease the mean rate of excretion and the observed extremes are shown in chart 3 in terms of the percentage of the injected

dose of salt eliminated at the four routine intervals, thirty, sixty, one hundred and twenty and one hundred and eighty minutes after the injection. As previously observed, the most conspicuous impairment exists in the first hour of the test. In the 80 cases of hypertensive

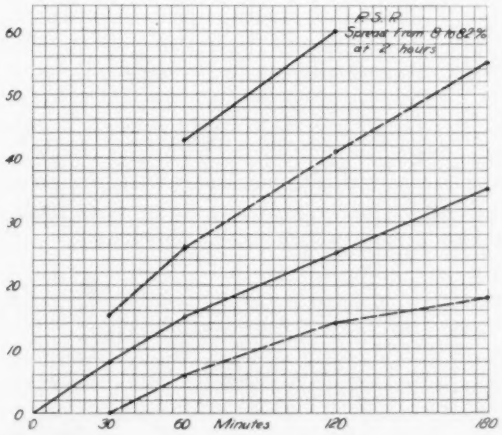


Chart 3.—Excretion of ferrocyanide in 80 cases of hypertensive disease of the arteries. The solid line represents the mean; the dash line, the extremes observed. The values are based on percentages of the injected dose.

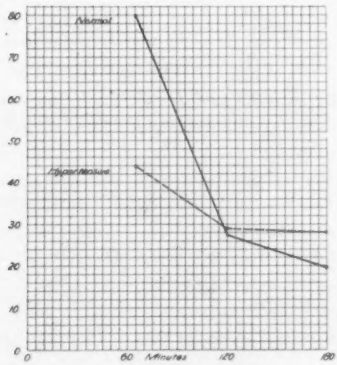


Chart 4.—Milligrams of ferrocyanide excreted by normal and by hypertensive patients (means).

arterial disease the average delay in excretion in the first hour is clearly seen in the curve when chart 3 is compared with the normal curve in chart 1. In the third hour the rate of excretion is much the same in hypertensive as in normal patients; if recorded in milligrams of

sodium ferrocyanide excreted (chart 4), the excretion of hypertensive patients in the last hour is actually greater than that of normal patients.

Analysis of the average excretion of sodium ferrocyanide of the 80 hypertensive patients by "correcting" the percentage of elimination for the amount of ferrocyanide already excreted reveals most interesting data (chart 5). Whereas for patients with the normal type of response the percentage of salt eliminated is at first high and then gradually declines, for hypertensive patients the curve of the mean percentage eliminated is a *rising* line. This rate is almost uniform throughout the three hour test. The spread of difference between it and the normal rate gradually diminishes, until in the third hour the rates are almost identical, the mean normal excretion being 10.41 per cent of the ferrocyanide remaining within the body and the mean excretion of patients with hypertensive arterial disease being 10.2 per cent.

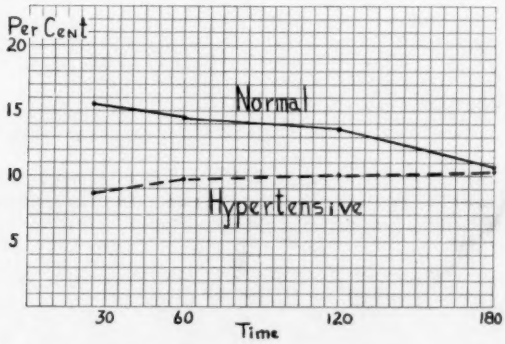


Chart 5.—Percentages of ferrocyanide excreted by normal and by hypertensive patients (means), "corrected" on the basis of the ferrocyanide remaining within the body.

Before considering the probable significance and interpretation of these observations on renal function in hypertensive disease of the arteries, one must point out that these data were obtained from patients with so-called "essential hypertension," with little or no evidence of renal injury. Patients with obvious nephritis and "secondary hypertension" and those with long-standing hypertensive arteriosclerosis with nephrosclerotic changes were rigidly ruled out as not representing the entity under consideration.[14] Efforts to find some correlation between the levels of arterial tension and the excretion of ferrocyanide were unavailing. No such correlation could be elicited in relation to the

14. Stieglitz, E. J.: (*a*) Arterial Hypertension, New York, Paul B. Hoeber, Inc., 1930; (*b*) Abnormal Arterial Tension, New York, National Medical Book Company, 1935; (*c*) J. Michigan M. Soc. **34**:70 (Feb.) 1935.

systolic or the diastolic tensions or the pulse pressures in individual patients or in groups of patients. In the series studied the systolic tensions ranged from 130 to 250 mm. of mercury, the diastolic tensions from 90 to 170 mm. and the pulse pressures from 35 to 105 mm. There was no relation between the ferrocyanide output and age. In all but a few instances the urine was clinically free of protein, although occasional hyaline and/or granular casts were frequently observed. In many instances the only evidence of renal injury was revealed by the test for impaired glomerular function with sodium ferrocyanide.

What is the significance of the conspicuous delay in elimination of ferrocyanide in hypertensive arterial disease and the almost uniform rate of excretion throughout the three hours of the test? In view of the present knowledge of the mechanisms of renal excretion,[14b] a tentative conclusion becomes obvious. Apparently, hypertensive disease of the arteries distinctly impairs the capacity of the glomeruli to respond to increased effort. Excretion of sodium ferrocyanide is a test for glomerular efficiency. The excretion of this salt is delayed and impaired in hypertension even though the urine may be apparently normal. Throughout the period of elimination studies, the percentage of salt excreted in relation to that remaining in the body is almost the same. This is most suggestive that in hypertensive disease the glomeruli are working at or near the maximum of their capacity all the time. That such impairment is revealed more readily and more clearly by tests with sodium ferrocyanide than by those with phenolsulfonphthalein and/or urea in the early stages of hypertensive disease is to be expected when one recalls the evidence demonstrating that ferrocyanide salts are eliminated wholly or almost wholly via the glomeruli whereas phenolsulfonphthalein [15] and urea likewise, in part,[16] are secreted by the proximal convoluted tubules. Injury to the glomeruli interferes with filtration and excretion more than with the free flow of blood.[17] The presence or absence of proteinuria is no criterion of the functional capacity of the kidneys. This view accords with the results of the epoch-making investigations of Goldblatt and his co-workers on the production of experimental hypertension by interference with the renal circulation.[18] Thus the present observations give further, clinical evidence that the glomeruli are vitally important factors in the pathogenesis of hypertensive arterial disease. The concept that glomerular elimination of water is impaired has been suggested as an etiologic mechanism in hypertension, but it appears

15. Potter, A. C., and Bell, E. T.: Am. J. M. Sc. **149**:236, 1915.

16. Stieglitz, E. J.: M. Clin. North America **21**:281 (Jan.) 1937; footnote 14 *b*.

17. Weiss, S.; Parker, F., Jr., and Robb, G. P.: Ann. Int. Med. **6**:1599 (June) 1933.

18. Goldblatt, H.; Lynch, J.; Hanzal, R. F., and Summerville, W. W.: J. Exper. Med. **59**:347 (March) 1934.

far more logical to presume impairment in the elimination of some solute or solutes whose normal route of excretion, like that of ferrocyanide, is via the glomeruli.[19]

As the test of glomerular function with sodium ferrocyanide approximately parallels the response elicited with the test of urea clearance [12] and the phenolsulfonphthalein test,[1] the rates of excretion of ferrocyanide for the first half hour were compared with the maximum value obtained for the specific gravity of the urine in the concentration test. The results are shown in chart 6. The routine of Fishberg [20] was employed for the concentration test, as it has yielded the most consistently satisfactory results with a maximum of simplicity in the procedure.[21] The renal concentration test is unquestionably considered

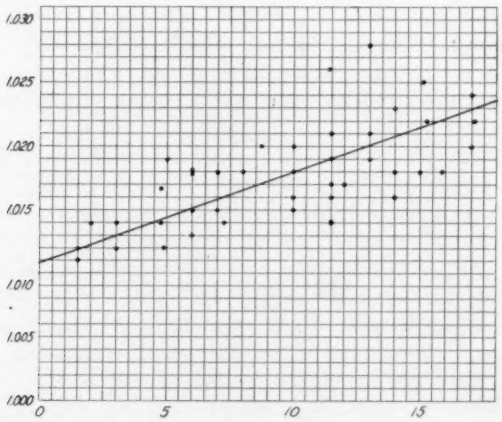


Chart 6.—Relation of the test of glomerular function to the maximum value for specific gravity obtained by concentration tests; ferrocyanide output during the first half-hour in terms of percentages of the injected dose.

the most sensitive indicator of early depreciation of the renal reserve, for it creates conditions of stress. It is physiologic, simple, safe and informative, and, although it measures primarily tubular effectiveness in concentrating the urine, it must be emphasized that functional impairment of the kidney *always involves both the tubules and the glomeruli.*[20] Glomerular injury inevitably impairs the tubular blood supply; tubular injury can come only via the blood stream, which has traversed the glomerular tufts before reaching the tubules. Thus it is

19. Stieglitz.[16] Goldblatt, Lynch, Hanzal and Summerville.[18]

20. Fishberg, A. M.: Urinary Nature of the Impairment of Renal Function, Arch. Int. Med. **38**:259 (Aug.) 1926; Hypertension and Nephritis, ed. 2, Philadelphia, Lea & Febiger, 1934.

21. Stieglitz (footnotes 14 *a* and *b* and 16).

truly impossible that either "pure" glomerular nephritis or "pure" tubular nephrosis exists; the functional impairment is always mixed, although the injury to one or the other structure may predominate. Thus, despite the fact that excretion of sodium ferrocyanide may be taken as a measure of glomerular efficiency, there should be and is a fairly close parallelism between the response elicited with all the tests of renal function.

As has been noted, the greatest deviation from the normal excretion of ferrocyanide occurs in the first hour after injection not only in hypertensive arterial disease but in acute and in chronic nephritis as well. It is therefore possible to shorten the procedure and analyze only specimens obtained thirty and sixty minutes after the injection, omitting the specimens at one hundred and twenty and one hundred and eighty minutes. For routine clinical purposes, especially with ambulatory patients, this shortening of the time required has been found entirely satisfactory. In prenatal evaluation of renal function [22] it is particularly appreciated by the patients, and there is no loss in the informativeness of the results. In a considerable but uncompleted series of prenatal observations the one hour application of the sodium ferrocyanide test has been useful warning of renal injury in a number of cases.

CONCLUSIONS

The essential criteria of a practical functional test of clinical value are simplicity, safety, specificity for the structure or function studied, creation of stress and relative uniformity of normal findings. The test of glomerular function with sodium ferrocyanide fulfils these criteria. But so do other tests. What then, if any, are the advantages offered by this method of measuring renal efficiency? Other than the far more cumbersome and technically difficult studies of creatinine or inulin clearance, the ferrocyanide test is thus far the only clinical method of specifically measuring the excretory activity of the glomeruli. The general utility of the test is attested by the parallelism of the responses to those elicited with the renal concentration test, the urea clearance studies and the phenolsulfonphthalein test. The rate of excretion is unaffected by the volume of urine secreted. In certain conditions of glomerular injury, as in the notable example of hypertensive arterial disease, the test of glomerular function reveals otherwise hidden impairment. In the present investigations it has been shown that in hypertensive disease without other evidence of renal injury the rates of excretion of ferrocyanide take on a characteristic form which implies definite change in the ability of the glomeruli to excrete solutes. These observations not only are compatible with but lend support to the con-

22. Stieglitz, E. J.: Obstetric Medicine, edited by F. L. Adair and E. J. Stieglitz, Philadelphia, Lea & Febiger, 1930, chap. 28.

cept that a primary initiating factor in the pathogenesis of hypertensive disease may be glomerular impairment. Further studies along these lines are needed and are contemplated.

The test is not applicable in cases of urinary obstruction due to prostatic hypertrophy and/or with urethral lesions, because of the urethral irritation set up. Utilization of the test in prenatal study offers specific advantages in the detection of lowered renal reserve, particularly in the discovery of preexistent hypertension and/or nephritis.[22]

SUMMARY

Sodium ferrocyanide, being excreted by the glomeruli, may be safely used as a test substance for determining glomerular efficiency.

The procedure found most applicable is the intravenous injection of 0.5 Gm. of the hydrated salt (representing 0.25 Gm. of the anhydrous salt) freshly dissolved in 10 cc. of sterile distilled water; specimens of urine are collected thirty, sixty, one hundred and twenty and one hundred and eighty minutes after injection and their ferrocyanide content determined. For ambulatory patients analysis of the first two specimens suffices. Caution must be employed in that the solution *must* be clear before the injection is made.

The excretion rate of ferrocyanide approximately parallels the results obtained with the urea clearance test, the phenolsulfonphthalein test and the renal concentration test.

Extension of the control series of patients with apparently normal renal function has further clarified what constitutes a normal response. The range of variation is not excessive (chart 1).

Calculating the rates of excretion on the basis of the ferrocyanide remaining within the body rather than the total dose of salt reveals that the rate of excretion is remarkably uniform throughout the period of study (chart 2).

In hypertensive arterial disease without other evidence of nephritis the glomerular function is early and clearly impaired. This depression is most conspicuously manifest during the first hour of excretion. The curve of the percentages of excretion in hypertensive disease is characteristic (chart 5) and suggests that occult glomerular injury depreciates the capacity of the glomeruli to excrete certain solutes promptly. The etiologic relations of these phenomena to hypertensive disease are briefly considered.

The final evaluation of the usefulness and clinical applicability of the test of glomerular function with sodium ferrocyanide can come only after extended investigation, but the present report clearly confirms the impressions gained in the preliminary studies and emphasizes its potential value in clinical study of the relation of glomerular injury to hypertensive disease.

RENAL FUNCTION AND THE NUMBER OF GLOMERULI IN THE HUMAN KIDNEY

J. M. HAYMAN JR., M.D.

J. W. MARTIN JR., M.D.

AND

MAX MILLER, M.D.

CLEVELAND

The majority of tests of renal function are interpreted and their results recorded in terms of the values obtained for normal persons. Any one of them yields an estimate of the efficiency of the kidneys at a particular time, but this may be of little prognostic significance, for it is well known that in acute nephritis, acute infections, ureteral obstruction or cardiac failure renal function may be severely depressed and yet with recovery of the patient may return to normal. In chronic renal disease, however, decreasing functional values usually indicate the relentless progress of the condition.

The ideal test of function would give not only an estimate of the degree of loss of renal efficiency but information as to the manner in which the normal physiologic processes have been disturbed. The processes involved in the formation of urine are so complex that no test has been devised by which it is possible satisfactorily to separate interference with glomerular function and interference with tubular function. Efforts to translate the results of tests of renal function into terms of renal physiology are, however, by no means new. From a comparison of urea clearance values and autopsy observations Van Slyke and his co-workers [1] suggested that in cases of hemorrhagic and degenerative nephritis this test is a measure of the number of intact glomeruli still functioning, while in cases of arteriosclerotic nephritis it is proportional

The expense of this study was defrayed in part by a grant from the Commonwealth Fund.

From the Department of Medicine, Western Reserve University, and the Medical Service of Lakeside Hospital.

A preliminary report of some of these observations was presented at the Fifty-Second Session of the Association of American Physicians, May 5, 1937 (Tr. A. Am. Physicians **52**:182, 1937).

1. Van Slyke, D. D.; Rhoads, C. P.; Hiller, A., and Alving, A. S.: Relationships Between Urea Excretion, Renal Blood Flow, Renal Oxygen Consumption, and Diuresis: The Mechanism of Urea Excretion, Am. J. Physiol. **109**:336 (Aug.) 1934.

to the decrease in renal blood flow. In both hemorrhagic and arteriosclerotic Bright's disease there is a marked reduction in the number of glomeruli.[2] The extent of such reduction may be difficult to judge from histologic sections, since the scars of destroyed glomeruli disappear without leaving recognizable traces. Taylor and his co-workers [3] presented evidence, based on studies of animals of various sizes, that the "urea ratio," a value derived in a manner similar to that in which the maximum clearance is determined, can be interpreted as a measure of the mass of functioning renal tissue. It is decreased after unilateral nephrectomy and increases again with compensatory hypertrophy of the remaining kidney. This interpretation offers difficulties in cases of temporary depression of function if it is asked what part of the renal mass has ceased to function. In rabbits poisoned with a uranium compound Watanabe, Oliver and Addis [4] grouped the lesions as mild, moderate and severe and found the urea ratio correspondingly depressed. This has been interpreted by others [5] to mean that the ratio is a measure of the mass of "functioning" renal tissue even when the total mass is not reduced. This conception of the "mass of functioning tissue" has been criticized by Cope.[6] Smith, Goldring and Chasis,[7] on the other hand, expressed the belief that the inulin, phenol red and diodrast clearances represent measures of glomerular filtrate, "tubular excretory mass" and effective renal blood flow. Elliot and Nuzum [8] concluded that it is difficult to interpret the results of tests of renal function in arteriosclerotic Bright's disease in terms of the actual amount of healthy renal parenchyma, a conclusion which led Schlayer [9] years ago to turn to functional studies in an effort to learn the kind rather than the amount of renal damage.

---

2. Moritz, A. R., and Hayman, J. M., Jr.: The Disappearance of Glomeruli in Chronic Kidney Disease, Am. J. Path. **10**:505 (July) 1934.

3. Taylor, F. B.; Drury, D. R., and Addis, T.: The Regulation of Renal Activity: VIII. The Relation Between the Rate of Urea Excretion and the Size of the Kidneys, Am. J. Physiol. **65**:55 (June) 1923.

4. Watanabe, C. K.; Oliver, J., and Addis, T.: Determination of the Quantity of Secreting Tissue in the Living Kidney, J. Exper. Med. **28**:359 (Sept.) 1918.

5. MacKay, E. M., and Rytand, D. A.: Significance of the Phenolsulphonphthalein Test of Renal Function, Arch. Int. Med. **55**:131 (Jan.) 1935.

6. Cope, C. L.: Rational Assessment of Renal Damage, Lancet **2**:799 (Oct. 13) 1934.

7. Smith, H. W.; Goldring, W., and Chasis, H.: The Measurement of the Tubular Excretory Mass, Effective Blood Flow and Filtration Rate in the Normal Human Kidney, J. Clin. Investigation **17**:263 (May) 1938.

8. Elliot, A. H., and Nuzum, F. R.: Evaluation of Measures of Renal Function in Persons with Arteriosclerotic Bright's Disease, Arch. Int. Med. **57**: 1151 (June) 1936.

9. Schlayer, C.: Neuere klinische Anschauungen über Nephritis, Med. Klin. (supp.) **8**:211, 1912; cited by Watanabe, Oliver and Addis.[4]

Tests of function may be interpreted from another point of view, namely, as measures of different renal functions without regard to the physiologic processes involved. Alving and Van Slyke [10] suggested that the specific gravity test determines chiefly the ability to excrete inorganic salts, while the urea clearance test measures the ability to excrete urea. Cope [6] concluded that all clearance tests simply measure the ability to eliminate waste and that the excretion of water and chloride is too dependent on extrarenal factors to be of value.

In order to attempt to analyze in terms of renal physiology the ways in which reduction in function may be brought about, it is necessary to know not only the results of the tests and the histologic changes in the kidneys but also the number and state of the glomeruli. This report deals with a comparison of the results of two commonly used tests, the urea clearance test and the maximum specific gravity or concentration test, and of one less widely employed, the creatinine clearance test, with the histologic appearance and particularly with the number of glomeruli.

The tests were performed from a few days to several months before death. All the patients had normal or elevated blood pressure and were free from detectable edema or other evidence of heart failure at the time the tests were performed. It is believed that by this restriction patients in whom reduced function could be attributed to diminished renal blood flow have been excluded. No patient with acute glomerulonephritis (in whom histologic evidence and the results of perfusion experiments also indicated the presence of diminished renal blood flow) was included. It was not possible, however, to carry out all three tests on every patient.

Urea clearance was calculated as "maximum" clearance and recorded as the percentage of normal for the observed volume of urine, the formula derived by Dominguez [11] as fitted to Van Slyke's data being used. The values were not corrected for surface area, since all the patients were adults and the correction was less than the variation in clearance values from one period to another. One to six urea clearance tests, each for two one hour periods, were made for each patient. Patients unable to cooperate in complete voiding were catheterized. The urea content of the urine was determined by the urease manometric method of Van Slyke and that of the blood by the same method or by the hypobromite method.[12]

10. Alving, A. S., and Van Slyke, D. D.: The Significance of Concentration and Dilution Tests in Bright's Disease, J. Clin. Investigation **13**:969 (Nov.) 1934.

11. Dominguez, R.: On the Renal Excretion of Urea, Am. J. Physiol. **112**:529 (July) 1935.

12. Peters, J. P., and Van Slyke, D. D.: Quantitative Clinical Chemistry, Baltimore, Williams & Wilkins Company, 1932, vol. 2.

Creatinine clearance was estimated by Rehberg's [13] method after administration of 3 to 5 Gm. of creatinine by mouth unless the creatinine content of the blood was above 5 mg. per hundred cubic centimeters. The values are expressed directly in cubic centimeters per minute. Hayman, Halsted and Seyler [14] found the average normal creatinine clearance to be 148 cc. per minute by this technic.

The concentration test used was a modification of the test of Addis and Shevky [15] and of Lashmet and Newburgh.[16] No fluid was allowed from 10 p. m. one evening until 7 a. m. thirty-three hours later, and the specific gravity of the urine passed during the last twelve hours of the test was determined. During the day of the test the diet recommended by Lashmet and Newburgh was used. This is palatable and adequate and diminishes thirst. It was not possible, however, to carry out this rigid regimen with all patients. In some cases twenty-four or twelve hours without fluids was all that could be reasonably asked. The thirty-three hour tests and the others are indicated by different symbols in the charts. Values for specific gravity were determined by means of a Westphal balance at 20°/4° with a calibrated plummet. All values for specific gravity were corrected for protein. This was determined by Shevky and Stafford's [17] method after calibration of the volume of precipitate by macro-Kjeldahl analyses.

Kidneys were obtained as soon after death as possible and perfused first with salt solution to wash out the blood and then with a mixture of ferric ammonium citrate and potassium ferrocyanide. The number of glomeruli per kidney was estimated by Kunkel's [18] method, with the modification that several blocks were cut before digestion, and the completeness of injection was estimated from counts of injected and apparently patent but noninjected glomeruli in sections. This permitted the estimate from the count to be corrected for the uninjected and therefore uncounted glomeruli. Except when there were gross infarcts, the distribution of uninjected glomeruli was usually fairly uniform. One

13. Rehberg, P. B.: Ueber die Bestimmung der Menge des Glomerulusfiltrats mittels Kreatinin als Nierenfunktionsprüfung, Zentralbl. f. inn. Med. **1**:367 (April) 1929.

14. Hayman, J. M., Jr.; Halsted, J. A., and Seyler, L. E.: A Comparison of the Creatinine and Urea Clearance Tests of Kidney Function, J. Clin. Investigation **12**:861 (Sept.) 1933.

15. Addis, T., and Shevky, M. C.: A Test of the Capacity of the Kidney to Produce a Urine of High Specific Gravity, Arch. Int. Med. **30**:559 (Nov.) 1922.

16. Lashmet, F. H., and Newburgh, L. H.: Improved Concentration Test of Renal Function, J. A. M. A. **99**:1396 (Oct. 22) 1932.

17. Shevky, M. C., and Stafford, D. D.: A Clinical Method for the Estimation of Protein in Urine and Other Body Fluids, Arch. Int. Med. **32**:222 (Aug.) 1923.

18. Kunkel, P. A.: The Number and Size of the Glomeruli in the Kidneys of Several Mammals, Bull. Johns Hopkins Hosp. **47**:285 (Nov.) 1930.

kidney from each patient was perfused, the other being subjected to the usual histologic examinations. Specimens in which there was a marked difference in weight or gross appearance of the two kidneys were discarded. Many other specimens were discarded because of failure to get good injection of the kidney. The two kidneys of a normal animal (rat,[19] rabbit [20] and man [21] contain approximately the same number of glomeruli. The average in the normal human kidney is about 1,250,000.

Seventy-nine patients were studied. Nineteen showed no evidence of renal disease during life, and no change or only slight change was found in the kidneys on histologic examination. These patients constituted a control group. The causes of death for this group were: miliary tuberculosis, 7; pneumonia, 4; a malignant process, 2, and lymphatic leukemia, duodenal ulcer, endocarditis lenta, Hodgkin's disease, coronary thrombosis and cerebral hemorrhage, 1 each. Urea clearance was determined for 16 patients in this group, creatinine clearance for 15 and specific gravity for 11. There were 2 patients with subacute and 7 with chronic glomerulonephritis. Urea clearance was determined for 8, creatinine clearance for 6 and specific gravity for 7. Thirty-nine patients had arteriolar nephrosclerosis of varying degree. Urea clearance was determined for 34, creatinine clearance for 31 and specific gravity for 29. Twelve patients had low renal function during life, but on postmortem examination only cloudy swelling or "bile nephrosis" and a normal number of glomeruli were found. Urea and creatinine clearances were determined for all of this group, specific gravity for only 3.

The patients showing reduced values for renal function may be divided into two groups: (1) those with chronic renal disease, either inflammatory or vascular, but without clinical evidence of acute infection; and (2) those with acute infections (typhoid, pneumonia), septicemia or obstructive jaundice but without chronic Bright's disease. The mechanism for the reduction in function in these two groups may be considered separately. The same 19 "normal" patients have been used as controls for both groups.

CHRONIC BRIGHT'S DISEASE (INFLAMMATORY AND VASCULAR)

When the mean creatinine or urea clearance is plotted against renal weight the correlation is extremely poor (chart 1). While in this study patients with kidneys weighing less than 100 Gm. each always showed

19. Arataki, M.: On the Postnatal Growth of the Kidney with Special Reference to the Number and Size of the Glomeruli, Am. J. Anat. **36**:399 (Sept.) 1926.

20. Hayman, J. M., Jr., and Starr, I.: Experiments on the Glomerular Distribution of Blood in the Mammalian Kidney, J. Exper. Med. **42**:641 (Nov.) 1925.

21. Moore, R. A.: The Total Number of Glomeruli in the Normal Human Kidney, Anat. Rec. **48**:153 (Jan.) 1931.

reduced clearance, extremely low clearance was always encountered in patients whose kidneys weighed 150 to 200 Gm. each. With kidneys weighing 100 to 175 Gm. each, the clearance values showed no correlation with renal weight. Nor in this adult population is the correlation any better if clearance is plotted against renal weight per square meter of body surface.[22]

This lack of correlation between clearance values and renal mass is in sharp contrast to Addis' and MacKay's data on normal animals and man. The size of the kidney, however, is no good measure of the

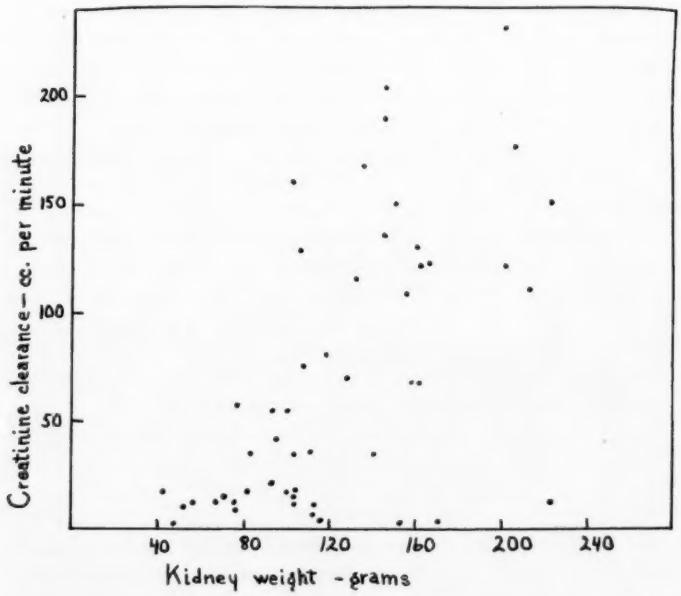
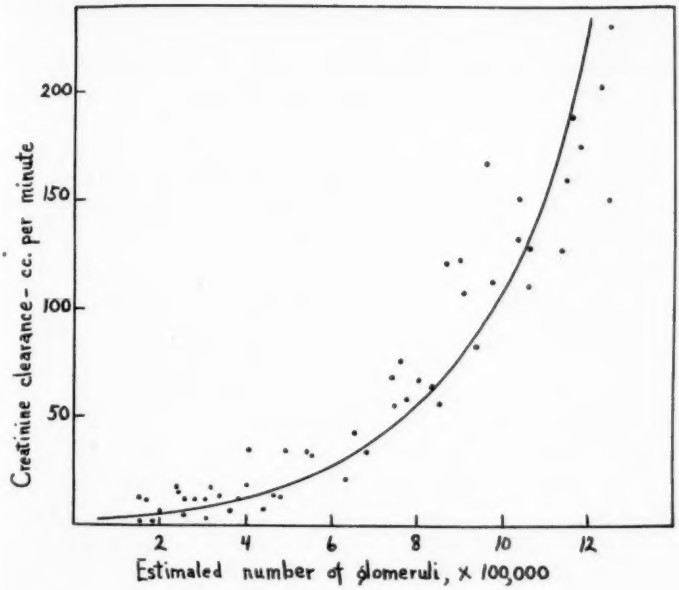


Chart 1.—Relation of average creatinine clearance and renal weight.

number of glomeruli it contains. Not only does the infant kidney have as many glomeruli as the adult kidney, but a relatively small pathologic adult kidney may have a fairly large number of glomeruli, while a kidney of approximately normal size, if it contains much scar tissue, may give a low count.

If, however, the mean urea or creatinine clearance value is plotted against the number of glomeruli per kidney, a reduction in clearance

22. MacKay, E. M.: Kidney Weight, Body Size and Renal Function, Arch. Int. Med. **50**:590 (Oct.) 1932.



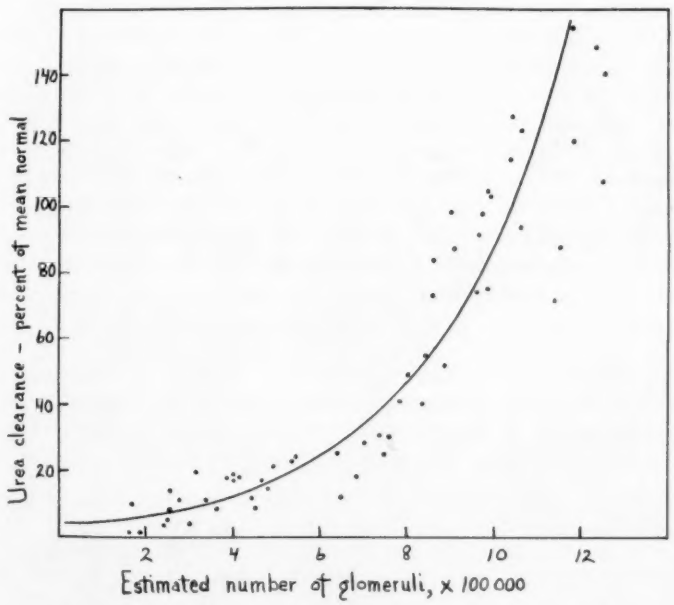


Chart 2.—Relation between creatinine and urea clearance and the estimated number of glomeruli per kidney in patients without evidence of renal disease and patients with chronic Bright's disease.

with a decreasing number of nephrons is apparent (chart 2). The relation, however, is not direct, clearance being reduced more rapidly than the number of nephrons. There is considerable scattering, but the points fall fairly well along an exponential curve. The curve of best fit has been calculated by the method of least squares. The curves for urea and for creatinine clearance are almost the same. The equation for creatinine is $Y = 3.186e^{0.00357x}$ and for urea $Y = 3.549e^{0.00322x}$, in which Y represents the clearance in cubic centimeters per minute for creatinine and in per cent of mean normal for urea, and X the estimated number of glomeruli per kidney. These equations are merely empiric correlations of observed data and obviously have no physiologic significance. This agrees with Hayman, Halsted and Seyler's conclusion that in chronic Bright's disease the percentage reduction below normal of urea and creatinine clearance is approximately the same.

In dogs in which the renal mass has been reduced by subtotal nephrectomy the percentage reduction in clearance is less than the reduction in glomeruli, so that the curve is convex upward.[23] In addition, the reduction in urea clearance is somewhat less than in creatinine clearance until the number of remaining glomeruli is extremely small. In these dogs two factors apparently contributed to the disproportionately large clearance in relation to renal mass. Glomerular counts include all glomeruli and are not necessarily a measure of the number through which blood was flowing or which were "active" when the clearance was determined. With moderate reduction in the number of glomeruli, it seems reasonable to suppose that all or the greater number of those which remain will be continuously active.[24] When renal mass has been reduced to a point where retention of nitrogen begins, it seems reasonable to suppose that all of the remaining glomeruli will be continuously open and that the relatively high clearance for the number of remaining glomeruli is attributable to a greater blood flow per glomerulus than occurs in the normal kidney.

Similar reasons may be advanced for the proportionately smaller reduction in clearance in relation to the number of glomeruli in the human kidney after unilateral nephrectomy. Contrary to the common belief that, owing to compensatory hypertrophy, renal function is normal after removal of half the renal mass, analysis of data presented by Ellis and Weiss[25] and similar data from this laboratory indicate that there is

23. Hayman, J. M., Jr.; Shumway, N. P.; Dumke, P., and Miller, M.: Experimental Hyposthenuria, J. Clin. Investigation **18**:195 (March) 1939.

24. Moore, R. A., and Lukianoff, G. F.: The Effect of Unilateral Nephrectomy on the Total Number of Open Glomeruli in the Rabbit, J. Exper. Med. **50**:227 (Aug.) 1929.

25. Ellis, L. B., and Weiss, S.: The Renal Function in Persons with One Kidney, Am. J. M. Sc. **186**:242 (Aug.) 1933.

a significant reduction in the mean creatinine and urea clearance.[26] The reduction, however, is not proportional to the reduction in renal mass but is similar in degree to that found in experimental animals.

While in experimental animals subjected to subtotal nephrectomy the remaining glomeruli are supposedly normal, those in human kidneys which are the seat of chronic Bright's disease are not. Consequently, the area of filtering surface will be reduced more than the number of glomeruli. By the method of counting, however, this was neglected, since a glomerulus which contained any dye was included. In an effort to correct for this defect Dr. A. R. Moritz and Dr. William Wartman of the staff of the Institute of Pathology studied sections of these kidneys and made an estimate of the degree of damage in from 100 to 200 consecutive glomeruli. Without knowledge of the results of the functional tests or of the glomerular counts, they grouped the glomeruli according to their appearance in the histologic sections into four categories, depending on whether they appeared to be normal or to be slightly, moderately or severely damaged. If it is assumed that the glomeruli in these four groups had, for instance, 100, 75, 50 and 25 per cent of the filtering surface of normal glomeruli, it is possible to derive a figure from the observed glomerular count which perhaps gives a closer approximation to the actual filtering area. With these figures, which are expressed in terms of an equivalent number of normal glomeruli, the plot of clearance against the calculated equivalent number of normal glomeruli still follows a curve which is convex downward, although less markedly so than when the observed number of glomeruli is used. If, however, it is assumed that the area of filtering surface in the four groups approximated 100, 50, 25 and 10 per cent of normal, the curves more nearly approach a straight line (chart 3). Such assumptions and calculations are, of course, purely arbitrary and in themselves are of no significance. They do, however, suggest that physiologic impairment in a glomerulus may be much greater than the degree of damage estimated from histologic sections. The fact that the rate of reduction in

26. Ellis and Weiss determined the renal function of 9 patients who had had a nephrectomy for tumor, stone or pyelonephrosis and who showed a normal blood pressure and no complications. Their figure for the mean creatinine clearance of normal persons was 151 cc., with a standard deviation of 13.6 cc., while the mean creatinine clearance for these apparently normal persons with one kidney was 114.0 ± 13.1 cc. The difference in the means of 37.0 ± 4.2 cc. is significant. We have examined 11 similar patients with one kidney. The mean creatinine clearance was 100.5 ± 3.4, which also differs significantly from the mean of 148 ± 34 obtained for normal persons by Hayman, Halsted and Seyler (difference of means equals 48 ± 4.6 cc.).

clearance in relation to the number of glomeruli falls more rapidly in chronic Bright's disease than after unilateral nephrectomy in man or subtotal nephrectomy in experimental animals is indirect evidence of a fundamental difference between the two processes. In the one the remaining nephrons are normal, in the other pathologic. The pathologic changes may include not only a reduction in area of filtering surface but a decrease in the permeability of the glomerular membrane, so that there is less filtrate for a given filtering area and capillary pressure. This is perhaps brought out more clearly if the average creatinine

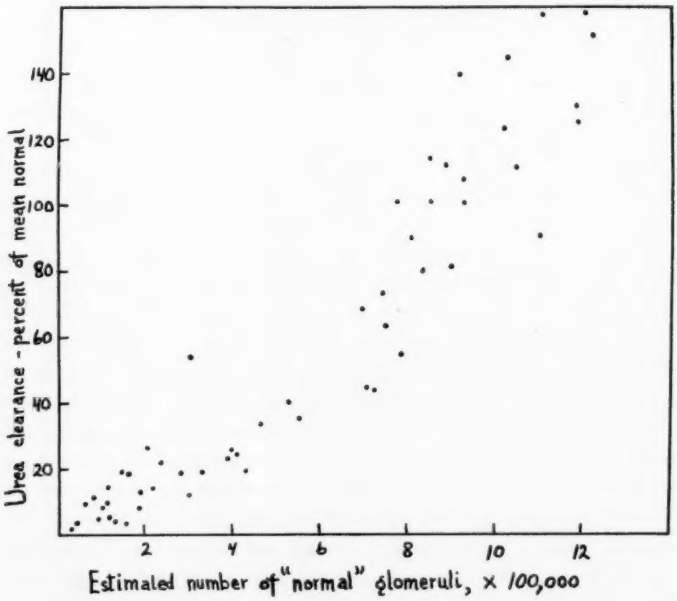


Chart 3.—Relation between urea clearance and the estimated number of "normal" glomeruli on the assumption that histologic grading represented 100, 50, 25 and 10 per cent of the normal filtering surface.

clearance per hundred thousand glomeruli is calculated. With two normal kidneys, if all the glomeruli are active this is about 5.9 cc. per hundred thousand, and with 700,000 glomeruli per kidney, about 2.9 cc. When the glomeruli are reduced to 200,000 per kidney the average clearance is only 1 cc. per hundred thousand. Comparable figures are obtained for urea clearance. The disproportionate reduction in clearance could, of course, also be accounted for if the tubule cells of the

remaining nephrons in these kidneys were damaged so that secretion of creatinine was reduced and back diffusion of urea increased to the same degree. There is nothing in these data to suggest that the aglomerular tubules in such kidneys described by Oliver and Luey [27] and by MacNider,[28] are functionally significant, at least in the elimination of urea and creatinine.

The concentration test is a sensitive, reliable and easily performed test of renal function, but it is well known that the specific gravity may reach a minimum value of about 1.010 while the patient is still free from

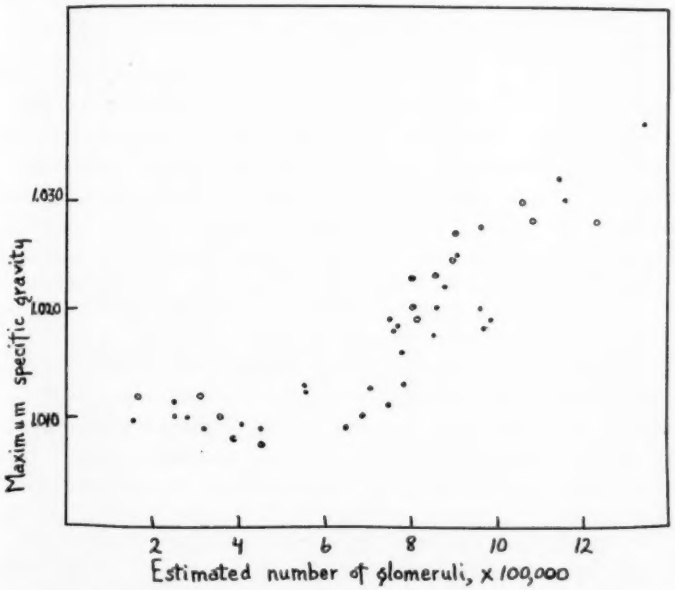


Chart 4.—Relation between the maximum specific gravity and the number of glomeruli per kidney in patients without evidence of renal disease and patients with chronic Bright's disease. The circles represent thirty-three hour concentration tests; the dots, those after shorter periods of abstinence from fluids.

symptoms and that with further progress of the disease no further reduction in specific gravity occurs. Alving and Van Slyke compared

27. Oliver, J., and Luey, A. S.: Plastic Studies in Abnormal Renal Architecture: Aglomerular Nephrons of Terminal Hemorrhagic Bright's Disease, Arch. Path. **19**:1 (Jan.) 1935.

28. MacNider, W. deB.: Pathological Changes in the Dog Kidney Resembling Normal Histological Structure in the Aglomerular Fish Kidney, Opsanus Tau, Proc. Soc. Exper. Biol. & Med. **31**:293 (Nov.) 1933.

the urea clearance and concentration tests and found that both were equally good until the specific gravity had fallen to 1.010, which occurred when the value for urea clearance had reached about 35 per cent of the average normal. After this the clearance test alone showed the progress of the disease.

These facts are confirmed by a correlation of the maximum specific gravity and the number of glomeruli (chart 4). The shape of the curve is different from that afforded by a correlation of the clearance value and the number of glomeruli. It is approximately a horizontal line up to between 700,000 and 800,000 glomeruli per kidney and then rises at an angle of 45 degrees to the normal range. The point of apparent discontinuity, about 700,000 or 800,000 glomeruli per kidney, corresponds to about 35 per cent of normal urea or creatinine clearance on the other curve. The reason for the distribution in this scatter diagram invites speculation. Van Slyke and his associates [29] found that the urea clearance was reduced to 40 per cent of the mean normal before there was any retention of urea. These data include only 4 patients with glomerular counts above 800,000 in whom the urea nitrogen content of the blood was above 20 mg. per hundred cubic centimeters, and the counts for all 4 of these were between 800,000 and 900,000. Only 2 patients with counts below 800,000 had a urea nitrogen content below 20 mg. per hundred cubic centimeters and only 4 below 30 mg., and the glomerular counts of these patients all ranged about 700,000. The inference seems strong that the apparent discontinuity corresponds to the point at which all of the remaining glomeruli become continuously active. The general shape of the scatter diagram is not changed if the maximum value for specific gravity is plotted against an equivalent number of "normal" glomeruli as described. The point of discontinuity then corresponds to about 400,000 to 500,000 glomeruli per kidney, which again corresponds roughly with a urea clearance of 35 per cent if one assumes that the area of filtering surface in the normal, slightly damaged, moderately damaged and severely damaged glomerulus is 100, 50, 25 and 10 per cent of that in a normal glomerulus.

There is little correlation between the systolic blood pressure and the estimated number of glomeruli, except that no patient was encountered with less than 700,000 glomeruli per kidney who had a systolic pressure below 150 mm. of mercury (chart 5). On the other hand, a

29. Van Slyke, D. D.; McIntosh, J. F.; Möller, E.; Hannon, R. R., and Johnston, C.: Studies of Urea Excretion: VI. Comparison of the Blood Urea Clearance with Certain Other Measures of Renal Function, J. Clin. Investigation **8**:357 (April) 1930.

person with a normal number of injectable glomeruli per kidney may have an elevated blood pressure. Below 700,000 glomeruli per kidney there was no correlation between the number of injectable glomeruli and the degree of elevation of blood pressure. Of the 7 patients with a glomerular count above 800,000 and a systolic blood pressure above 140 mm., all but 1 excreted urine with a specific gravity above 1.020 as ascertained by the concentration test, and this one showed a gravity of 1.019. While this may be a coincidence and dependent on the small

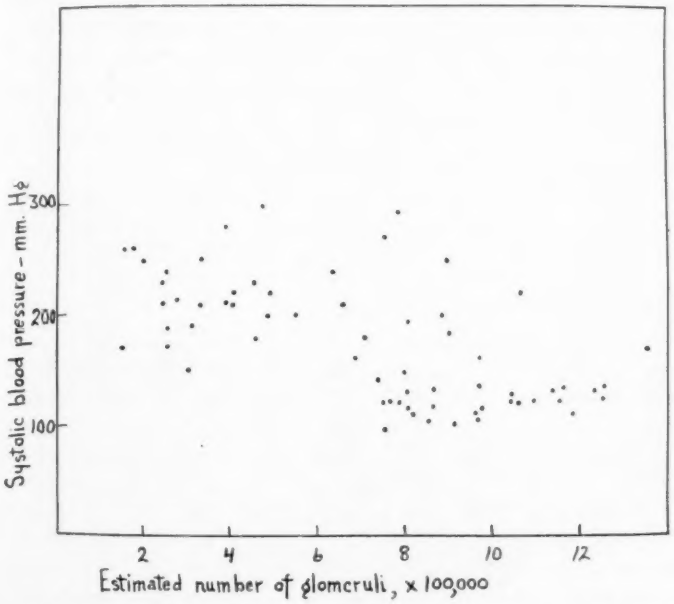


Chart 5.—Relation between the systolic blood pressure and the number of glomeruli per kidney.

number of observations, the data at hand at least suggest that when the glomeruli have been reduced to 700,000 or 800,000 per kidney there is some change associated with the presence of elevated blood pressure and loss of concentrating power. It does not follow, however, that these are causally related, for dogs in which Goldblatt and his co-workers [30] had produced marked persistent hypertension by constriction of the

30. Goldblatt, H.; Lynch, J.; Hanzal, R. F., and Summerville, W. W.: Studies on Experimental Hypertension, J. Exper. Med. **59**:347 (March) 1934.

renal arteries retained a normal concentrating power, and some dogs in which hyposthenuria had been produced by subtotal nephrectomy showed no significant rise in blood pressure.[28]

TOXIC NEPHROSES

Dogs poisoned with uranium acetate showed marked reduction in clearance and inability to concentrate their urine in spite of a normal number of glomeruli and a normal blood pressure and renal blood flow.[23] This is most readily attributed to damage to the tubule cells, which

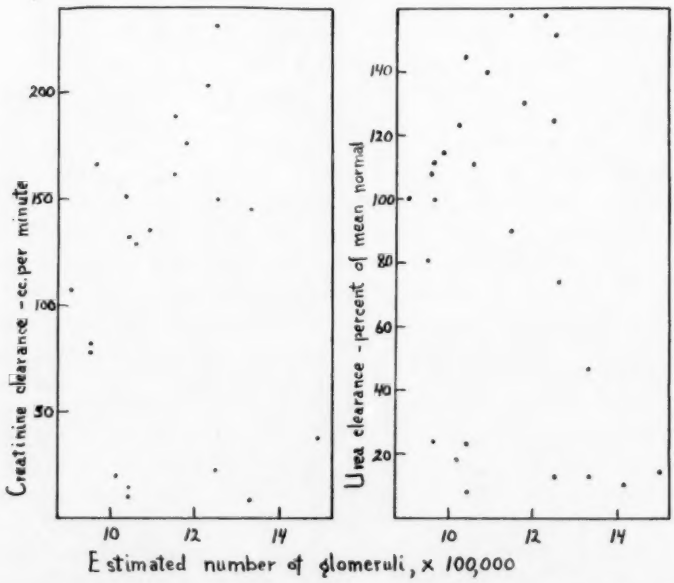


Chart 6.—Relation between creatinine and urea clearance and the estimated number of glomeruli per kidney in patients without evidence of renal disease and patients with toxic nephroses.

interferes with the reabsorption of water and at the same time permits more back diffusion not only of urea but of creatinine. Similarly, some patients with such acute infections as pneumonia and typhoid or with miliary tuberculosis, septicemia or jaundice show markedly reduced clearance in spite of a normal number of glomeruli which histologically show little or no damage and a normal blood pressure (chart 6). If the patient recovers from the original disease the clearance returns toward normal.

Data on concentrating power are lacking because withholding fluids for a long period was not justified. In chronic renal disease the specific gravity of the urine just before death is usually at least as high as that obtained with a concentration test, provided an infusion has not been given recently. If such specimens represent approximately maximal concentrating ability, then such data as are available indicate that these patients are unable to excrete a urine of normally high specific gravity.

It is, of course, obvious that a combination of these two mechanisms, reduction in the number of nephrons and tubular damage, leading to low clearance may occur. When a patient with chronic renal disease suffers an acute infection or a ureteral obstruction, clearance may suddenly be markedly reduced and may then return toward the previous level when the infection or other cause of depression disappears. Other mechanisms causing low clearance may be diminished renal blood flow or an inadequate pressure in the glomerular capillaries. The former may occur in acute nephritis and in cardiac failure; the latter, in shock, hemorrhage or Addison's disease. Patients in whom any of these conditions (or combinations of them) were present have, as far as possible, been excluded from this study.

SUMMARY

Creatinine and urea clearance, maximum urinary specific gravity and blood pressure were correlated with the number of glomeruli per kidney, estimated after postmortem perfusion, in patients with and in patients without disease of the kidneys.

In chronic glomerulonephritis and nephrosclerosis the values for creatinine and urea clearance are closely correlated with the number of glomeruli. Maximum specific gravity falls with the decrease in the number of glomeruli until the latter reaches 700,000 to 800,000 per kidney, after which it remains fixed in spite of further reduction in the number of glomeruli. If the number of glomeruli per kidney is less than 700,000, the systolic blood pressure is invariably above 150 mm.

In certain cases of acute infections and jaundice, clearance and concentrating ability may both be markedly reduced in spite of a normal number of glomeruli showing no significant changes in histologic sections.

BACTERIAL ENDOCARDITIS

REPORT OF A CASE IN WHICH THE CAUSE WAS ACTINOMYCES BOVIS

NATHANIEL UHR, M.D.
NEW YORK

The clinical syndrome of subacute bacterial endocarditis occurs frequently and is today a well recognized entity. Little has been added to knowledge of the subject since Libman's publications.[1] It is now well known that while Streptococcus viridans is the most common cause of this disease, a large variety of other bacteria may produce a similar picture. Among these are Bacillus influenzae, Enterococcus, Meningococcus, Bacillus diphtheriae, Pneumococcus, Gonococcus and organisms of the Brucella group. However, the occurrence of a similar clinical picture caused by higher bacteria is rare. Only 1 case—that of Jervell,[2] in which the condition was caused by Leptothrix—is to be found in the literature. For this reason, the case to be described is of unusual interest, since the causative agent of the disease was found to be Actinomyces bovis.

REPORT OF CASE

History.—W. J., a 24 year old American-born Jew, a chiropodist, was admitted to the medical service of the Hospital for Joint Diseases on Oct. 16, 1936. The family history was noncontributory. The only disease he had had as a child was measles. There was no history of rheumatism, chorea or sore throats. In 1931 he had been observed at another hospital for an attack of acute epigastric pain, associated with fever and acholic stools but without jaundice (?), and had improved in a few days, without any diagnosis being made. The condition never recurred.

On Oct. 5, 1936, he became ill with pain in both calves, malaise, nausea and vomiting. Thereafter for six days he had a spiking temperature of 104 F. in the morning, attended by chills and sweats. For the next four days the temperature remained at an approximate level of 102 F.

From the Medical Service of the Hospital for Joint Diseases; Dr. A. A. Epstein, Director.

1. Libman, E., and Celler, H.: The Etiology of Subacute Infective Endocarditis, Am. J. M. Sc. **140**:516, 1910. Libman, E.: Cardiac Lesions of Subacute Bacterial Endocarditis, ibid. **164**:313, 1912; The Clinical Features of Subacute Streptococcus (and Influenzal) Endocarditis in the Bacterial Stage, M. Clin. North America **11**:117, 1918. Libman, E., and Friedberg, C. K.: Bacterial Endocarditis, in Piersol, G. M.; Borty, E. L., and others: Cyclopedia of Medicine, Philadelphia, F. A. Davis Company, 1932, vol. 3, p. 181.

2. Jervell, O.: Leptothrix in the Blood in a Case of Malignant Endocarditis, Norsk mag. f. lægevidensk. **83**:36, 1922.

Physical Examination.—Examination on admission revealed an acutely ill subject with a temperature of 104 F. Respiration was normal; the skin was clear; the eyegrounds were normal and the scleras ware not discolored. The heart was not demonstrably enlarged; the sounds were forceful, regular and of good quality; the rate was 120. A soft, short systolic murmur was localized at the apex. The lungs were clear. There were increased spasticity and some tenderness in the right upper quadrant of the abdomen, with tenderness on deep percussion over the lower right costal cage. The spleen was not palpable. There was no lymphadenopathy. There was no nuchal rigidity or other abnormal neurologic finding. No clubbing of the fingers or toes was noted. A tentative diagnosis of typhoid fever was made, although bacterial endocarditis was seriously considered.

Course.—During the remaining three and a half weeks of the patient's life, his fever fluctuated between 103 and 104 F. The faint systolic murmur heard on admission gradually became harsher, louder and more widely transmitted. Six days after admission he began to cough and expectorate blood-streaked sputum. Dulness and diminished respiratory sounds were found at the base of the right lung. The spleen seemed enlarged on percussion but still could not be felt. The patient's general condition became steadily worse, and on the eighth day after admission petechiae were first noted in the left conjunctiva and on the left arm. At this stage the diagnosis of subacute bacterial endocarditis was made. Dr. E. Libman, who saw the patient in consultation, concurred in this diagnosis. Slight cyanosis of the lips and finger nails appeared, and fine crepitant rales now were heard at the bases of both lungs. New crops of petechiae, many of which had pale centers, appeared daily on various parts of the body. In addition, somewhat larger purpuric lesions developed first over the palms and soles and later over other parts of the body. The sensorium gradually became clouded, and on the eighteenth day the patient lapsed into a stupor, which deepened progressively. Dyspnea became marked, and the signs in the chest increased. Gangrenous areas developed on the lobe of the right ear, both heels and the left leg. Coma became deeper; signs of left hemiplegia developed, and death occurred on November 10, twenty-five days after admission.

Treatment had consisted of the usual supportive measures, in addition to two transfusions and oxygen administered by nasal catheter. In view of the nature of the organism, the patient was given three doses of sodium iodide, 15 grains (0.97 Gm.), intravenously, but this treatment was discontinued in the absence of any favorable response.

Laboratory Data.—The urine was of good concentration throughout the illness; albumin was present in traces; sugar was absent; microscopic examination on various occasions revealed red blood cells, a few white blood cells and occasional hyaline and granular casts. Culture of the urine revealed no organisms. The initial erythrocyte count of 4,640,000, with 80 per cent hemoglobin (Sahli), fell to 3,200,000, with 50 per cent hemoglobin, before death. The leukocyte count gradually rose from 12,000, with 80 per cent polymorphonuclear neutrophils, to a peak of 38,000, with 93 per cent polymorphonuclears. The platelet count was 256,000. The sedimentation rate on November 6 was 41 mm. The Wassermann, Kahn and gonococcus fixation tests gave negative reactions. Agglutination tests for Bacillus typhosus, Bacillus paratyphosus A and B and Brucella abortus gave negative results. Agglutination with Bacillus proteus in a dilution of 1:40 was present. Examination of sputum on October 22 revealed no tubercle bacilli. The blood chemistry was normal on admission. On October 29 the icteric index was 17.4; on November 6, the urea nitrogen was 49.2 mg. per hundred cubic centimeters and the icteric index 16.7.

Roentgen examination of the lungs on October 17 gave normal results. On November 6 marked congestion at the bases of both lungs was reported. An electrocardiogram made on October 21 showed only sinus tachycardia. On November 6 the tracings showed sinus tachycardia, inversion of T_1, low voltage of T_2 and T_3 and left axis deviation.

Each of four cultures of the blood, taken consecutively at intervals over about three weeks, yielded a pure culture of A. bovis. Cultures required four to eight days for growth. The organism grew only in the presence of blood and was microaerophilic, requiring reduced oxygen tension for isolation. Morphologically gram-positive, nonacid-fast tangled threads of mycelium were observed, accompanied in older cultures by numerous abnormal forms. This strain was not pathogenic for rabbits (fig. 1).

Fig. 1.—Photomicrograph of a stained specimen of the blood culture.

Autopsy.—The body was that of a young man of average stature, somewhat emaciated, in complete rigor mortis. The eyes were sunken and the cheeks hollow. There was evident loss of subcutaneous fat. The skin over the face, chest, abdomen and extremities was studded with innumerable petechiae, many of which were white centered. Petechiae were evident also in the conjunctivas. Neither jaundice nor superficial lymphadenopathy was noted.

The panniculus adiposus was thin. The peritoneal surfaces were everywhere smooth and glistening. The small intestine showed occasional bluish-discolored, irregular flecks and contained a small amount of fluid. The liver was enlarged and extended below the costal margin, as did the spleen. The diaphragm was elevated and extended to the level of the fifth interspace on each side.

There were no pleural adhesions. There appeared to be a small amount of blood-tinged fluid in the right side of the chest, although the precise amount could not be ascertained because of the abdominal approach. The lungs were voluminous and heavy. They were crepitant throughout, except for several

circumscribed, bluish segments of atelectasis in the lower lobe posteriorly. The upper lobe of the right lung showed a protruding, circumscribed, firm, nodular lesion, about 1 cm. in diameter, with a reddish congested zone around it. On section of this lesion, a cluster of fairly firm, grayish, granular, slightly elevated areas was seen, suggesting small granulomatous nodules or abscesses rather than the ordinary type of bronchopneumonia. Several areas of similar appearance were encountered in the lungs, and sections were taken through these areas. On section, the lungs otherwise had a mottled dark red appearance and exuded frothy fluid on slight pressure. The bronchi also contained pinkish frothy material. There were no enlarged hilar lymph nodes. Microscopic section showed small confluent patches of bronchopneumonia, with exudate of polymorphonuclear leukocytes and apparently some fibrin in alveoli. Several bronchi in the field also contained

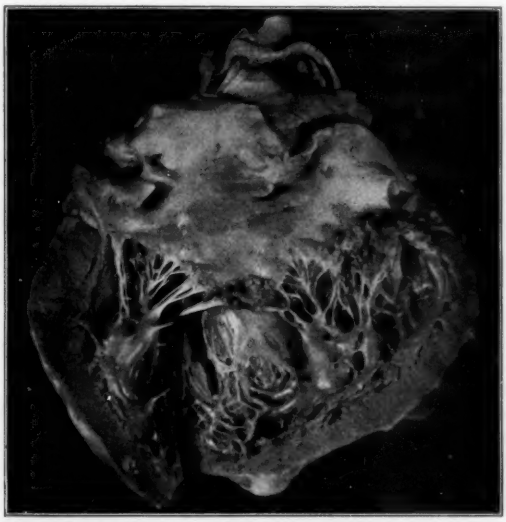


Fig. 2.—Section of the heart, showing vegetation on the mitral valve.

purulent exudate, and their walls were infiltrated by polymorphonuclear leukocytes; many brown pigment-containing phagocytes were noted within alveoli. Pulmonary edema was present.

The pericardial surfaces were smooth and glistening. The heart was enlarged and globular, owing to dilatation of both the right and the left ventricle. The myocardium was flabby, soft and pale yellowish brown. The tricuspid, the pulmonary and the aortic valve showed no gross evidences of inflammation. The mitral valve, however, showed a large, soft, friable, yellowish vegetative lesion, involving principally the aortic cusp but distributed also in small clusters along the remainder of the line of closure. The vegetations extended to the anterior and posterior surfaces of the cusps of the valve for some distance, but no destruction or perforation of the valve was noted anywhere (fig. 2). The mitral valve showed no evidence of previous valvulitis; no thickening or vascularization of the margin

of the valve was apparent, while the chordae tendineae were slender and delicate and inserted normally in the valve. There was also a small, circumscribed vegetative lesion on the endocardium of the left auricle, similar in appearance to the valvular lesion and situated about 1 cm. above it. There was no conspicuous thickening of the auricular endocardium which might suggest an old rheumatic auricular lesion. The papillary muscles and the ventricular endocardium showed nothing noteworthy. Section of the myocardium showed a few small, grayish-discolored areas suggesting embolic lesions, which were taken for section. Microscopic sections showed myocardial degeneration. There were scattered small areas of myocardial necrosis and infiltration with small mononuclear cells and polymorphonuclear leukocytes. Many of these inflammatory foci were perivascular.

The aorta showed scattered yellow intimal atheromatous patches in the descending portion.

The liver was large, soft and yellowish. The anterior surface of both the right and the left lobe showed fibrous perihepatitis and a fibrinous exudate of more recent origin. On section, the lobular outline could be recognized, as well as moderate central lobular congestion, severe parenchymatous degeneration and conspicuous fatty infiltration. The gallbladder and the bile ducts showed nothing abnormal and presented no evidence of infection. The portal vein appeared normal. Microscopic sections showed severe degeneration of liver cells, with vacuolation. The sinusoids were congested and the Kupffer cells enlarged. The periportal fields showed infiltration with mononuclear cells and some eosinophils.

The spleen was markedly enlarged, being approximately 9 to 10 inches (23 to 25 cm.) in length and extending about 2 fingerbreadths below the costal margin. It was fairly soft. The surface presented several circumscribed discolored infarcts, varying from 1 to 4 cm. in diameter. The pulp on section was dark red and presented prominent grayish nodules and streaks. The splenic artery and vein showed nothing unusual. Microscopic sections showed a number of small infarcts. The malpighian follicles were small. The pulp was hyperemic and contained a considerable number of macrophages and polymorphonuclear leukocytes. Much pigment, most of which was probably due to fixation in solution of formaldehyde, was observed; a bacterial clump was seen in an arteriole; the pulp contained many polymorphonuclear leukocytes, including eosinophils and macrophages. Sections also showed an infarct with a congested peripheral zone. No granulomatous or proliferative reaction was noted.

The kidneys were slightly enlarged and were normal in shape and position. The capsule stripped without difficulty, revealing a surface studded with many pinhead-sized reddish petechial lesions, apparently embolic in nature, as well as a number of small yellowish infarcts, about 0.5 cm. in diameter. There was rather severe parenchymatous degeneration. The markings of the cortex and medulla were indistinct. The pelvis likewise showed a considerable number of petechial hemorrhages. The ureters appeared normal. The urinary bladder was studded with multiple pinhead-sized petechiae, many of which were white centered. The prostate showed nothing noteworthy. Microscopic sections of the kidneys showed engorgement of interlobular arteries and their branches. The epithelium of the convoluted tubules presented severe degeneration with formation of vacuoles and hyaline droplets. Scattered tubular casts were noted. Focal areas showed interstitial infiltration with polymorphonuclear leukocytes and numerous mononuclear cells. No embolic focal lesions were demonstrated.

The adrenals showed a thin yellow cortex and an autolyzed medulla. Microscopic section showed nothing noteworthy.

The pancreas showed the usual lobular structure. Microscopic sections showed postmortem change.

The stomach showed prominent rugae and moderate congestion. The small intestine, when opened, showed innumerable embolic petechial lesions in the mucosa, many with white centers. Similar lesions were noted in even greater number in the colon, the sigmoid and the rectum. There were no conspicuously enlarged mesenteric lymph nodes.

The brain could not be examined, because the examination was limited. Portions of several lumbar vertebrae were taken for study of the bone marrow.

Bacteriologic Studies.—Culture of the crushings of the mitral vegetations revealed A. bovis.

Further studies on cultures submitted to the department of bacteriology of the United States Public Health Service confirmed the finding of A. bovis.

COMMENT

Ideal criteria for the diagnosis of bacterial endocarditis consist of a number of factors. The clinical picture should conform to the usual accepted standards; positive cultures of a single organism should be obtained from the blood during life, preferably prior to the last week of illness; the same organism should be recovered from the vegetations on the valves. These ideal demands are fulfilled in the case reported. The question whether the condition falls under the acute or the subacute variety need not be of concern here. While it is true that the duration of the illness was only thirty-five days, the case presented the characteristics of the ordinary case of subacute bacterial endocarditis. The use of any time, such as six weeks, as a basis for subdivision is arbitrary and without constant pathologic justification.

The case of Jervell assumes special importance in the present connection, since it is the only case besides mine in which one of the higher bacteria was found as a cause of the endocarditis. The patient was a 20 year old man whose illness began with gastrointestinal symptoms, followed by chills, fluctuating temperature and cutaneous hemorrhages. Embolization of the brain and spinal cord was striking. Death occurred eight weeks after the onset. Autopsy revealed old rheumatic mitral valvulitis, with fresh vegetations on both the aortic and the mitral valve and small ulcerations. The spleen was enlarged and showed many infarcts, some of which were abscessed. Cultures of the blood during life and vegetations of the valves showed Leptothrix.

The question of the relation of actinomycosis to cardiac involvement now presents itself. Actinomycosis of the heart is rare. Sanford and Voelker,[3] in an exhaustive statistical review of 670 cases of actinomycosis occurring in the United States, failed to find a single case in which the disease involved the heart. Nevertheless, such involvement

3. Sanford. A. H., and Voelker, M.: Actinomycosis in the United States. Arch. Surg. **11**:809 (Dec.) 1925.

is not unknown, and cases have been reported by Poncet and Berard,[4] Benda,[5] Harbitz and Gröndahl,[6] Letulle and Hufnagel,[7] Dean[8] and Kasper and Pinner.[9] In all of these cases, the pericardium and myocardium became involved by direct extension from the pleura and mediastinum or by metastatic spread through the blood stream from a distant focus. Dean's case and 1 of those of Harbitz and Gröndahl are particularly noteworthy, since endocardial vegetations were seen in addition to the myocardial involvement. In both cases the endocardial process was due to direct extension from an underlying myocardial lesion.

It is apparent, therefore, not only that endocarditis due to A. bovis is a rare complication of actinomycosis but that even when it does occur it is entirely different in its method of invasion and pathogenesis from ordinary bacterial endocarditis. Clinically, too, the primary disease dominates the picture.

In my case of endocarditis due to Actinomyces no portal of entry was demonstrated after a thorough search. The clinical picture was indistinguishable from that of ordinary subacute bacterial endocarditis, despite the relatively short duration of its course. Pathologically, too, none of the lesions distinctive of actinomycosis was seen. The case therefore derives its especial importance from two angles: first, the unusual form of involvement of the heart by Actinomyces and, second, the unique nature of this etiologic agent as a cause of typical bacterial endocarditis.

SUMMARY

A case of bacterial endocarditis due to A. bovis is reported. Clinically and pathologically it was indistinguishable from an ordinary case of subacute bacterial endocarditis.

The Pathological Department of the Hospital for Joint Diseases supplied the pathologic and microbiologic studies in the case and the photographs shown in figures 1 and 2.

145 West Fifty-Eighth Street.

4. Poncet, A., and Berard, L.: Traité clinique de l'actinomycose humaine, Paris, Masson & Cie, 1898.

5. Benda, C. E.: Zwei Fälle von metastasierenden Aktinomykose, Deutsche med. Wchnschr. **26**:70, 1900.

6. Harbitz, F., and Gröndahl, N. B.: Die Strahlenpilzkrankheit (Aktinomykose) in Norwegen, Beitr. z. path. Anat. u. z. allg. Path. **50**:193, 1911.

7. Letulle, M., and Hufnagel, L.: L'actinomycose du coeur, Bull. Acad. de méd., Paris **82**:120, 1919.

8. Dean, G.: A Case of Pyaemic Actinomycosis with an Actinomycotic Endocarditis, Brit. M. J. **2**:1303, 1912.

9. Kasper, J. A., and Pinner, M.: Actinomycosis of the Heart: Report of a Case with Actinomycotic Emboli, Arch. Path. **10**:687 (Nov.) 1930.

PROTAMINE ZINC INSULIN: A METABOLIC STUDY

TREATMENT IN TWO CASES OF SEVERE DIABETES BY EQUALLY AND UNEQUALLY DIVIDED DIETS, WITH COMMENTS ON CRITERIA FOR TREATMENT

EDWARD TOLSTOI, M.D.
AND
FREDERICK C. WEBER JR., M.D.
WITH THE TECHNICAL ASSISTANCE OF VINCENT TOSCANI, B.S.
NEW YORK

During the past two years we have treated some of our diabetic patients, both in the outpatient department and in the hospital wards, with protamine zinc insulin. Like others,[1] we hoped that by virtue of its slow activity a single daily dose of this new preparation could be substituted for three or even four doses of regular insulin. However, when we put this thought into practice we experienced considerable difficulty in maintaining the urine free from sugar. We appreciated the facts that we employed the higher carbohydrate diet—200 to 250 Gm.—and that we had not worked out a technic for the use of protamine insulin. As a matter of fact, there is no definitely established technic for the administration of this preparation. All sorts of combinations[2] are recommended, with or without the addition of regular insulin—that is, two or more hypodermic injections daily. It was, and still is our feeling that if advantage is to be taken of the slow-acting property of protamine insulin efforts should be directed to treatment of the diabetic patient by means of *one* dose daily. Such a procedure would relieve the patient

From the New York Hospital and the Department of Medicine, Cornell University Medical College, and the Russell Sage Institute of Pathology.

1. Adlersberg, D.: Wien. klin. Wchnschr. **49**:476, 1936. Kerr, R. B., and Best, C. H.: Canad. M. A. J. **34**:400, 1936. Krarup, N. B.: Clinical Investigations into the Action of Protamine Insulinate, translated by C. L. Heel, Copenhagen, G. E. C. Gad, Publisher, 1935. Lawrence, K. D., and Archer, N.: Brit. M. J. **1**:747, 1936. Rabinowitch, I. M.; Fowler, A. F., and Corcoran, A. C.: Canad. M. A. J. **35**:239, 1936. Root, H. F.; White, P.; Marble, A., and Stotz, E.: Clinical Experience with Protamine Insulinate, J. A. M. A. **106**:180 (Jan. 18) 1936. Sprague, R. G.; Blum, B. B.; Osterberg, A. E.; Keppler, E. J., and Wilder, R. M.: Clinical Observations with Insulin Protamine Compound, ibid. **106**:1701 (May 16) 1936. Keppler, E. J.: Clinical Experience with Protamine Zinc Insulin, J. A. M. A. **110**:92 (Jan. 8) 1938.

2. Tolstoi, E.: Am. J. M. Sc. **194**:727, 1937.

of the burdensome routine of carrying his medication about and eating his meals at definite intervals in relation to the administration of insulin. And, as Mosenthal [3] aptly stated:

> The resort to protamine zinc insulin should do away with these restrictions; the ideal to be attained is that one dose of protamine zinc insulin is given in the morning before breakfast when the routine preparations for the day are carried out, and the meals are taken at optional hours with no anxiety about their being served exactly on time. Any deviation from this schedule does not fulfil the complete purpose of protamine zinc insulin.

To attain this ideal with a liberal carbohydrate diet, various dietary divisions instead of the customary three equal portions of carbohydrate throughout the day have been recommended. The Danish discoverers [4] of protamine insulin were the first to suggest the irregular distribution of the carbohydrate fraction. S. Harris and S. Harris Jr.[5] recommended six meals in place of the usual three. Other investigators [6] advised that the day be started with a small carbohydrate meal and that some carbohydrate be given before bedtime. Another method was to fractionate the total carbohydrate into ⅕, ⅖, ⅖ for three meals and then subtract 10 to 15 Gm. of carbohydrate from each meal, to be given two to three hours after each meal. Pollack and Lande [7] have experimented with such dietary divisions, and they have no one fixed formula as yet. They used the method which gave them the best results, and in some cases it was a ⅕, ⅖, ⅖ division of the carbohydrate; in others it was ⅕, ⅕, ⅗, or ⅖, ⅕, ⅗. They further advised the omission at breakfast of foods containing immediately available sugar, such as fruit juices.

A perusal of the literature reveals that some or all of these methods are in use. It is also apparent that both the low and the high carbohydrate diet are employed. And, as there are no detailed data on record which deal with the response of the *same* diabetic patient to these various fractional dietary divisions or to a high or a low carbohydrate diet while one dose of protamine insulin is being received daily, we felt that a study of this type would be of interest. We set before

3. Mosenthal, H. O.: Protamine Zinc Insulin: Clinical Application, J. A. M. A. **110**:87 (Jan. 8) 1938.

4. Hagedorn, H. C.; Norman Jensen, B.; Krarup, N. B., and Wodstrup, I.: Protamine Insulinate, J. A. M. A. **106**:177 (Jan. 18) 1936.

5. Harris, S., and Harris, S., Jr.: Am. J. Digest. Dis. **5**:88, 1938.

6. Protamine Insulin and Diet in Diabetes Mellitus, editorial, Ann. Int. Med. **11**:2048, 1938.

7. Pollack, H., and Lande, H.: New York State J. Med. **38**:339, 1938. Pollack, H.: J. Mt. Sinai Hosp. **4**:437, 1938.

us the following three questions, the answers to which we hope may be of considerable help in the treatment of the diabetic patient.

1. Is glycosuria affected materially by shifting the diet from ⅓, ⅓, ⅓ to ⅕, ⅖, ⅖ if all other factors remain constant?

2. Is the patient's diabetes better managed with a lower than with a higher carbohydrate diet?

3. What are to be considered criteria for the "control" of diabetes?

PLAN OF EXPERIMENT

The subjects of our study were 2 "healthy" female diabetic women. Their diabetes was severe, as can be noted from their respective clinical histories, which will be given. During the eighteen months prior to the writing of this report neither was free from glycosuria, even though each received 50 to 80 units of insulin—protamine as well as regular. Prior to admission they adhered to their diets as well as they could. They are intelligent persons, and they made every effort to cooperate.

We hospitalized both patients, one for fifty and the other for sixty days. Both were under rigid supervision in the metabolism ward. Each was given a diet of 75 Gm. of protein, 60 Gm. of fat and 200 Gm. of carbohydrate, a total of 1,640 calories. As the protocols show, 1 of the patients was given 50 and the other 60 units of protamine zinc insulin daily before breakfast. The experiment was divided into ten day periods. During the first, the diet was fractioned into three approximately equal portions. During the second period the distribution of the diet was ⅕, ⅖, ⅖. We then resumed the original proportions of ⅓, ⅓, ⅓ for another ten days. At the end of these three periods the carbohydrate fraction was reduced by one-half, and to maintain the diet isocaloric the same number of calories was added to the fat. The new diet consisted of 75 Gm. of protein, 105 Gm. of fat and 100 Gm. of carbohydrate. The daily dose of insulin remained the same throughout the entire experiment, as did the quantity of protein. The figure of 100 for carbohydrate was not selected at random. We observed that our patients excreted about 100 Gm. of dextrose a day. This excretion was, of course, variable, but the total output for the first thirty days was 3,000 Gm. Since the patients failed to utilize this amount, we felt that a deduction of the daily average excretion—100 Gm.—from the original diet was reasonable. After the patients were given the lower carbohydrate diet, they again were observed when the division of the food was ⅛, ⅛, ¼, when it was ⅕, ⅖, ⅖ and when it was ⅖, ⅕, ⅖. The patients were not in bed all the time. They were seen daily and questioned as to symptoms of diabetes and as to their well-being in general. Their weight was recorded daily.

Every specimen of urine voided was preserved in a separate receptacle. Each one was tested for dextrose qualitatively. The specimens for a twenty-four hour period were then pooled, and, after the daily volume was noted, the total amount of dextrose as well as of nitrogen was determined. Tests for acetone also were done daily on the total twenty-four hour specimen.

On the eighth day of *each* period a blood sugar curve was recorded. This was taken during fasting and at 11 a. m., and 1, 4 and 7 p. m.

The Benedict qualitative and quantitative method was used for the determination of the urinary sugar. Acetone was determined by the Rothera test. The

Folin-Wu method was used for the analyses of blood sugar and the Bloor method for the estimation of cholesterol.

REPORT OF CASES

CASE 1.—F. H., a 27 year old single white woman, whose personal history and family history revealed no facts contributory to the present situation, about four years before noted continued hunger, thirst and marked loss of weight, in spite of eating a good deal and drinking large amounts of water. At that time she had to be fitted for glasses because of blurred vision. The menstrual periods became scantier during the three to four months these symptoms were present, and her weight dropped from 140 pounds (63.5 Kg.) to 114 pounds (52 Kg.). While treating her for a minor injury, a chiropractor elicited the aforementioned symptoms and on examining a specimen of her urine told her that she had diabetes and sent her to the clinic at Billings hospital in Chicago, in July 1934. Physical examination at that time revealed no abnormalities other than that the liver was felt at 2 cm. below the costal margin. Glycosuria (4 plus) was present, and the urine gave a 2 plus reaction for acetone. She was given a diet of 60 Gm. of protein, 175 Gm. of fat and 100 Gm. of carbohydrate, and after some treatment she was finally discharged on the same diet with insulin in doses of 16-6-12. On this regimen her weight rose to 130 pounds (59 Kg.) in January 1935. However she continued to have either insulin reactions or glycosuria. In January 1937, while in Florida, she contracted a cold, which was diagnosed as influenza. She was ill for about a month. Her weight dropped to 111 pounds (50.5 Kg.), and of course the glycosuria could not be regulated at that time.

On her return from Florida she came to the diabetic clinic of the New York Hospital, in May. On her admission the diet consisted of 60 Gm. of protein, 175 Gm. of fat and 100 Gm. of carbohydrate, with insulin in doses of 30-15-15. Glycosuria (4 plus) was present and the urine gave a 3 plus reaction for acetone. The results of the physical examination were in no way contributory. The diet was changed at this time to 70 Gm. of protein, 60 Gm. of fat and 200 Gm. of carbohydrate, with the dose of insulin as stated, 30-15-15. At almost every visit to the clinic she had glycosuria (4 plus) and a 0 to 2 plus reaction for acetone in the urine. She stated that the insulin reactions were troublesome. Since she seemed to be the type in which the blood sugar oscillates, it was decided to try protamine insulin, and in July she was given 40 units of protamine and 15 units of regular insulin before breakfast. On this regimen she felt better than she ever did before. Her weight rose to 134 pounds (60 Kg.), but she continued to have irregular glycosuria. As she continued to have occasional reactions, the dose was reduced to 35 units of protamine and 10 units of regular insulin in the morning. From August to February 1938 she felt well and took care of herself. She was excreting sugar all the time during her absence. She reported to the hospital in February because of this constant glycosuria and an infection of the gum. In view of the fact that she had severe diabetes which was difficult to regulate, she was requested to enter the metabolism ward for study.

On her admission the physical examination revealed nothing abnormal except for a slight infection of the gum, which was treated. The urine revealed sugar (4 plus) but no albumin or red blood cells. The erythrocyte count was 4,900,000, with 94 per cent hemoglobin, and the leukocyte count 8,200, with a normal differential count. The blood sugar content during fasting was 214 mg. per hundred cubic centimeters. The blood showed no serologic abnormality, and the basal metabolic rate was + 7 per cent.

CASE 2.—M. D., an 18 year old white girl, who has been followed closely for the past six years, was first seen in April 1932, at the age of 12 years, during a bout of diabetic ketosis necessitating admission to the hospital. A paternal grandfather died of diabetes at 65. The child grew and developed normally. About March 1932 she lost 12 pounds (5.4 Kg.) in two weeks' time; she was hungry and voided large amounts of urine. She became irritable and lethargic about six days before admission. She began to have epigastric pain and vomiting, and since the abdominal pain persisted she was brought to the clinic. At that time there was a marked odor of acetone on her breath. She felt dry and was sleepy. The urine had a 4 plus reaction for sugar and a 4 plus reaction for acetone. The sugar content of the blood was 625 mg. per hundred cubic centimeters. She was admitted to the hospital, where she responded satisfactorily to treatment and was discharged on a diet of 85 Gm. of protein, 110 Gm. of fat and 120 Gm. of carbohydrate, with insulin in doses of 10-0-10.

After her first admission she was in the hospital ten times. At no time after the discovery of her diabetes had her urine been sugar free, either at the hospital or at home. All sorts of dietary combinations as well as doses of insulin have been tried, but at no time was there continuous freedom from glycosuria. Whenever the dose of insulin was increased she was brought to the hospital because of profound shock. In spite of this difficulty, she continued to develop and grow. Her menses became established in 1935. In April of that year she was admitted to the hospital because of otitis media and acute mastoiditis, due to the beta hemolytic streptococcus. Mastoidectomy was done, and she made a good recovery, after which she was discharged on a diet of 70 Gm. of protein, 50 Gm. of fat and 250 Gm. of carbohydrate, with insulin in doses of 40-20-20. She continued to excrete sugar on this regimen, and the dose of insulin was changed to 30-20-20-10. Because some reactions occurred on this regimen the dose was reduced to 20-15-15-5. She began to gain weight and "felt fine," but the diet and the dose of insulin had to be altered constantly because of difficulty in adjustment. During this time her menses were irregular, and an estrogenic preparation (amniotin) was given, 2,000 to 10,000 units every other day for a month, but without success.

In June 1938 protamine insulin was administered in the evening, with regular insulin in the morning and at noon, and though the patient felt well she continued to excrete sugar. Thus she was treated, as has been described, by means of every conceivable combination of diet and insulin, but at no time could one obtain the laboratory ideal of satisfactory therapy. In view of the fact that she continued to develop normally, gain weight and height, and carry on in a perfectly normal manner in school, leading her class, in spite of constant glycosuria, less and less attention was being paid to that condition.

When she was admitted to the hospital, about April 1938, because of a slight infection of the upper part of the respiratory tract, it was decided to place her in the metabolism ward for study. A diet of 75 Gm. of protein, 60 Gm. of fat and 200 Gm. of carbohydrate was prescribed and one dose of protamine insulin given daily. The laboratory data other than those concerned with the diabetes were of no importance.

ANALYSIS OF DATA

At the outset we wish to emphasize that during the entire period of observation both patients felt perfectly well. Neither revealed a single symptom of diabetes. As can be seen in the tabulated data, F. H. lost

TABLE 1.—*Data for Patient F. H.*

Date	Weight, Kg.	Diet per 24 Hr.					Protamine Insulin, Units	Urine per 24 Hr.				
		Protein, Gm.	Fat, Gm.	Carbohydrate, Gm.	Calories	Division of Diet		Amount	Sugar, Gm.	Sugar, %	Total Nitrogen, Gm.	Nitrogen Balance*
2/28/38	60.6	49	41	124	1,095	1/3, 1/3, 1/3	40 (10 regular insulin)	2,365	104	4.4	9.18	+1.62
3/ 1/38	59.73	75	60	199	1,687	1/3, 1/3, 1/3	50	2,420	156.5	6.5	10.5	+0.3
3/ 2/38	59.6	75	59	200	1,640	1/3, 1/3, 1/3	50	2,210	125	5.6	10.75	+0.05
3/ 3/38	59.7	75	60	200	1,640	1/3, 1/3, 1/3	50	2,510	191	7.6	11.3	−0.5
3/ 4/38	59.68	75	60	199	1,640	1/3, 1/3, 1/3	50	2,275	193	8.5	10.71	+0.09
3/ 5/38	59.6	75	59	199	1,640	1/3, 1/3, 1/3	50	1,985	152	7.7	10.9	−0.1
3/ 6/38	59.2	75	60	199	1,640	1/3, 1/3, 1/3	50	1,540	96	6.2	10.75	+0.05
3/ 7/38	59.38	75	59.8	199	1,640	1/3, 1/3, 1/3	50	2,015	132	6.5	11.25	−0.45
3/ 8/38	59.19	75	60	200	1,640	1/3, 1/3, 1/3	50	1,495	90.9	6	11.3	−0.5
3/ 9/38	59.46	75	60	200	1,640	1/3, 1/3, 1/3	50	2,430	144	5.9	12.9	−2.1
3/10/38	58.85	75	60	200	1,610	1/3, 1/3, 1/3	50	1,430	154	10.7	10.55	+0.25
3/11/38	58.92	75	60	200	1,640	1/5, 2/5, 2/5	50	1,090	41.8	3.8	9.7	+1.1
3/12/38	58.94	75	60	200	1,640	1/5, 2/5, 2/5	50	1,395	59	4.2	9.7	+1.1
3/13/38	58.92	75	60	200	1,640	1/5, 2/5, 2/5	50	1,595	143	9	10.3	+0.5
3/14/38	58.59	75	60	200	1,640	1/5, 2/5, 2/5	50	1,745	143	8.2	11.14	−0.34
3/15/38	58.46	75	60	200	1,640	1/5, 2/5, 2/5	50	930	41	4.4	9.32	+1.48
3/16/38	58.82	75	60	200	1,640	1/5, 2/5, 2/5	50	1,040	31.3	3	9.35	+1.45
3/17/38	58.84	75	60	200	1,640	1/5, 2/5, 2/5	50	1,515	111	7.3	9.65	+1.15
3/18/38	58.82	75	60	200	1,640	1/5, 2/5, 2/5	50	1,265	51.8	4.1	9.7	+1.1
3/19/38	58.96	75	60	200	1,640	1/5, 2/5, 2/5	50	1,420	30.8	2.2	9.35	+1.45
3/20/38	58.67	75	60	200	1,640	1/5, 2/5, 2/5	50	1,285	45.5	3.5	9.03	+1.77
3/21/38	58.87	75	60	200	1,640	1/5, 2/5, 2/5	50	1,550	63.3	4.1	9.64	+1.16
3/22/38	58.4	75	60	200	1,640	1/5, 1/3, 1/3	50	1,500	125	8.3	10.3	+0.5
3/23/38	58.73	75	60	200	1,640	1/3, 1/3, 1/3	50	975	30.8	3.1	8.63	+2.17
3/24/38	59.25	75	60	200	1,640	1/3, 1/3, 1/3	50	1,620	83.5	5.1	9.82	+0.98
3/25/38	58.91	75	60	200	1,640	1/3, 1/3, 1/3	50	1,135	14.3	1.2	9.25	+1.55
3/26/38	58.89	75	60	200	1,640	1/3, 1/3, 1/3	50	1,440	61.5	4.2	9.93	+0.87
3/27/38	58.85	75	60	200	1,640	1/3, 1/3, 1/3	50	1,730	143	8.2	11.6	−0.8
3/28/38	58.89	75	60	200	1,640	1/3, 1/3, 1/3	50	1,040	83.5	8	9.36	+1.44
3/29/38	58.34	75	60	200	1,640	1/3, 1/3, 1/3	50	1,200	80	6.6	9.92	+0.88
3/30/38	58.51	75	60	200	1,640	1/3, 1/3, 1/3	50	1,505	100	6.5	10.63	+0.17
3/31/38	58.46	75	60	200	1,640	1/3, 1/3, 1/3	50	1,630	87	5.3	10.08	+0.72
4/ 1/38	58.48	75	105	100	1,640	1/3, 1/3, 1/3	50	865	2.6	0.3	9.37	+1.43
4/ 2/38	59	75	105	100	1,640	1/3, 1/3, 1/3	50	1,315	9.55	0.7	9.92	+0.88
4/ 3/38	58.82	75	105	100	1,640	1/3, 1/3, 1/3	50	1,170	8.35	0.7	6.22	+4.58
4/ 4/38	58.82	75	105	100	1,640	1/3, 1/3, 1/3	50	1,010	0	0	10.74	+0.06
4/ 5/38	58.92	75	105	100	1,640	1/3, 1/3, 1/3	50	1,210	14.3	1.2	10.52	+0.28
4/ 6/38	58.43	75	105	100	1,640	1/3, 1/3, 1/3	50	1,235	5.5	0.4	10.49	+0.31
4/ 7/38	58.42	75	105	100	1,640	1/3, 1/3, 1/3	50	925	0	0	9.53	+1.27
4/ 8/38	58.5	75	105	100	1,640	1/3, 1/3, 1/3	50	1,280	23.6	1.8	10.4	+0.4
4/ 9/38	58.32	75	105	100	1,640	1/3, 1/3, 1/3	50	1,105	40	3.6	10.02	+0.78
4/10/38	57.96	75	105	100	1,640	1/3, 1/3, 1/3	50	945	0	0	9.63	+1.17
4/11/38	58.22	75	105	100	1,640	1/3, 1/3, 1/3	50	945	0	0	9.75	+1.05
4/12/38	58.24	75	105	100	1,640	2/5, 1/5, 2/5	50	1,040	0	0	8.75	+2.05
4/13/38	58.19	75	105	100	1,640	2/5, 1/5, 2/5	50	1,255	37.2	2.9	11.92	−1.12
4/14/38	57.76	75	105	100	1,640	2/5, 1/5, 2/5	50	955	6.25	0.6	10.08	+0.72
4/15/38	58.31	75	105	100	1,640	2/5, 1/5, 2/5	50	860	0	0	9.92	+0.88
4/16/38	58.04	75	105	100	1,640	2/5, 1/5, 2/5	50	890	3.64	0.4	9.03	+1.77
4/17/38	57.87	75	105	100	1,640	2/5, 1/5, 2/5	50	950	0	0	10.63	+0.17
4/18/38	58.48	75	105	100	1,640	2/5, 1/5, 2/5	50	1,515	0	0	10.35	+0.45
4/19/38	58.44	75	105	100	1,640	2/5, 1/5, 2/5	50	1,455	16.4	1.1	9.58	+1.22
4/20/38	57.93	75	105	100	1,640	2/5, 1/5, 2/5	50	850	5	0.6	9.14	+1.66
4/21/38	58.07	75	115	125	1,895	1/3, 1/3, 1/3	50	1,185	18.9	1.6	10.3	+0.5
4/22/38	58.37	75	115	125	1,895	1/3, 1/3, 1/3	50	955	12.5	1.3	9.65	+1.15

Date	Weight, Kg.	Diet per 24 Hr.					Protamine Insulin, Units	Urine per 24 Hr.				
		Protein, Gm.	Fat, Gm.	Carbohydrate, Gm.	Calories	Division of Diet		Amount	Sugar, Gm.	Sugar, %	Total Nitrogen, Gm.	Nitrogen Balance*
4/24/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
4/25/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
4/26/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
4/27/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
4/28/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
4/29/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
4/30/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/ 1/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/ 2/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/ 3/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/ 4/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/ 5/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/ 6/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/ 7/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/ 8/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/ 9/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/10/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/11/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/12/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/13/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/14/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/15/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/16/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/17/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/18/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/19/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/20/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/21/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/22/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/23/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/24/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/25/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/26/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/27/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/28/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/29/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/30/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17
5/31/38	58.26	75	115	125	1,895	1/3, 1/3, 1/3	50	1,000	0	0	9.63	+1.17

Date	Weight, Kg.	Diet per 24 Hr. Protein, Gm.	Fat, Gm.	Carbohydrate, Gm.	Calories	Division of Diet	Protamine Insulin, Units	Urine per 24 Hr. Amount	Sugar, Gm.	Sugar, %	Total Nitrogen, Gm.	Nitrogen Balance*
4/20/38	57.93	75	105	100	1,640	1/3, 1/3, 1/3	50	1,185	18.9	1.6	10.5	+0.35
4/21/38	58.07	75	115	125	1,895	1/3, 1/3, 1/3	50	935	12.5	1.3	9.65	+1.15
4/22/38	58.37	75	115	125	1,895	1/3, 1/3, 1/3	50					
4/24/38	50.36	75	60	200	1,640	1/3, 1/3, 1/3	60	3,715	301.5	8.1	11.6	−0.8
4/25/28	49.18	75	60	200	1,640	1/3, 1/3, 1/3	60	2,480	156.2	6.3	10.35	+0.45
4/26/38	48.28	75	60	200	1,640	1/3, 1/3, 1/3	60	1,725	83.5	4.8	8.57	+2.23
4/27/38	48.92	75	60	200	1,640	1/3, 1/3, 1/3	60	1,620	80	4.9	9 25	+1.55
4/28/38	49.30	75	60	200	1,640	1/3, 1/3, 1/3	60	925	21.8	2.3	6.95	+3.85
4/29/38	49.57	75	60	200	1,640	1/3, 1/3, 1/3	60	1,485	66.8	4.5	8.68	+2.12
4/30/38	49.43	75	60	200	1,640	1/3, 1/3, 1/3	60	1,480	58.8	3.9	8.75	+2.05
5/ 1/38	49.3	75	60	200	1,640	1/3, 1/3, 1/3	60	1,360	71.5	5.2	9.13	+1.67
5/ 2/38	49.25	75	60	200	1,640	1/3, 1/3, 1/3	60	1,455	83.5	5.7	8.58	+2.22
5/ 3/38	49.28	75	60	200	1,640	1/3, 1/3, 1/3	60	1,120	41.8	3.7	7.23	+3.57
5/ 4/38	49.83	75	60	200	1,640	1/5, 2/5, 2/5	60	1,945	143.5	7.4	9.29	+1.51
5/ 5/38	49.12	75	60	200	1,640	1/5, 2/5, 2/5	60	2,170	129	5.9	12.95	−2.15
5/ 6/38	49.63	75	60	200	1,640	1/5, 2/5, 2/5	60	1,345	64.5	4.8	9.92	+0.88
5/ 7/38	49.8	75	60	200	1,640	1/5, 2/5, 2/5	60	1,555	87	5.6	8.58	+2.22
5/ 8/38	49.8	75	60	200	1,640	1/5, 2/5, 2/5	60	1,925	110	5.7	10.65	+0.15
5/ 9/38	49.55	75	60	200	1,640	1/5, 2/5, 2/5	60	1,400	74.2	5.3	11.04	−0.24
5/10/38	49.65	75	60	200	1,640	1/5, 2/5, 2/5	60	2,025	107	5.3	10.85	−0.05
5/11/38	49.66	75	60	200	1,640	1/5, 2/5, 2/5	60	2,170	143.8	6.6	10.12	+0 68
5/12/38	49.65	75	60	200	1,640	1/5, 2/5, 2/5	60	1,675	111	6.6	8.53	+2.27
5/13/38	49.61	75	60	200	1,640	1/5, 2/5, 2/5	60	1,875	93.5	5	8.97	+1.83
5/14/38	49.59	75	60	200	1,640	1/3, 1/3, 1/3	60	1,940	95.3	4.9	9.92	+0.88
5/15/38	49.23	75	60	200	1,640	1/3, 1/3, 1/3	60	1,970	100	5.1	11.38	−0.58
5/16/38	49.14	75	60	200	1,640	1/3, 1/3, 1/3	60	1,675	87	5.2	11.1	−0.3
5/17/38	49.5	75	60	200	1,640	1/3, 1/3, 1/3	60	2,220	115	5.1	14.15	−3.35
5/18/38	49.95	75	60	200	1,640	1/3, 1/3, 1/3	60	1,625	93.5	5.7	11.2	−0.4
5/19/38	49.92	75	60	200	1,640	1/3, 1/3, 1/3	60	1,505	74	4.9	10.8	±0
5/20/38	49.86	75	60	200	1,640	1/3, 1/3, 1/3	60	2,655	136	5.1	12.63	−1.83
5/21/38	50.07	75	60	200	1,640	1/3, 1/3, 1/3	60	2,215	139	6.2	10.14	+0.66
5/22/38	49.83	75	60	200	1,640	1/3, 1/3, 1/3	60	1,030	67	6.5	6.93	+3.87
5/23/38	49.3	75	60	200	1,640	1/3, 1/3, 1/3	60	1,415	83.5	5.9	10.38	+0.42
5/24/38	49.64	75	105	100	1,640	1/3, 1/3, 1/3	60	940	28.6	3	9.35	+1.45
5/25/38	49.85	75	105	100	1,640	1/3, 1/3, 1/3	60	1,220	50	4.1	9.08	+1.72
5/26/38	49.33	75	105	100	1,640	1/3, 1/3, 1/3	60	1,775	87	4.9	12.1	−1.3
5/27/38	49.33	75	105	100	1,640	1/3, 1/3, 1/3	60	845	40	4.7	7.45	+3.35
5/28/38	48.66	75	105	100	1,640	1/3, 1/3, 1/3	60	1,280	69	5.4	9.42	+1.38
5/29/38	49.94	75	105	100	1,640	1/3, 1/3, 1/3	60	1,370	66.8	4.9	9.98	+0.82
5/30/38	49.5	75	105	100	1,640	1/3, 1/3, 1/3	60	1,650	83.3	5	11.2	−0.40
5/31/38	48.92	75	105	100	1,640	1/3, 1/3, 1/3	60	1,500	69	4.6	11.2	−0.40
6/ 1/38	48.61	75	105	100	1,640	1/3, 1/3, 1/3	60	1,055	19.7	1.9	9.98	+0.82
6/ 2/38	48.7	75	105	100	1,640	1/3, 1/3, 1/3	60	1,640	83.3	5.1	11.3	−0.5
6/ 3/38	48.48	75	105	100	1,640	1/5, 2/5, 2/5	60	2,045	91.7	4.4	9.42	+1.38
6/ 4/38	48.63	75	105	100	1,640	1/5, 2/5, 2/5	60	1,585	95.5	6	9.88	+0.92
6/ 5/38	49.25	75	105	100	1,640	1/5, 2/5, 2/5	60	1,205	74.2	6.1	8.58	+2.22
6/ 6/38	49.55	75	105	100	1,640	1/5, 2/5, 2/5	60	1,235	76	6.1	8.85	+1.95
6/ 7/38	49.68	75	105	100	1,640	1/5, 2/5, 2/5	60	1,615	91	5.6	11.35	−0.55
6/ 8/38	49.6	75	105	100	1,640	1/5, 2/5, 2/5	60	1,995	116.3	5.8	13.03	−2.23
6/ 9/38	49.49	75	105	100	1,640	1/5, 2/5, 2/5	60	1,450	78.2	5.4	10.46	+0.34
6/10/38	49.97	75	105	100	1,640	1/5, 2/5, 2/5	60	850	45.5	5.4	6.72	+4 08
6/11/38	49.68	75	105	100	1,640	1/5, 2/5, 2/5	60	2,075	115	5.5	11.1	−0.3
6/12/38	49.35	75	105	100	1,640	1/5, 2/5, 2/5	60	1,900	113.5	6 0	15.1	−4.3
6/13/38	49.23	75	60	200	1,640	1/3, 1/3, 1/3	60	2,235	140.5	6.3	12.4	−1.6
6/20/38	48.97	75	60	200	1,640	1/3, 1/3, 1/3	60	1,990	117.5	5.9	10.87	−0.07
6/21/38	49.66	75	60	200	1,640	1/3, 1/3, 1/3	60	2,235	151	6.7	11.05	−0.25
6/22/38	49.49	75	60	200	1,640	1/3, 1/3, 1/3	60	1,870	97.3	5.2	10.4	+0.4
6/23/38	49.68	75	60	200	1,640	1/3, 1/3, 1/3	60	1,945	105	5.4	9.34	+1.46
6/24/38	49.2	75	60	200	1,640	1/3, 1/3, 1/3	60	2,015	137.5	6.8	10.5	+0.3
6/25/38	49.19	75	60	200	1,640	1/3, 1/3, 1/3	60	1,640	95.5	5.8	9.52	+1.28
6/26/38	49.15	75	60	200	1,640	1/3, 1/3, 1/3	60	2,455	116	4.7	10.82	−0.02
6/27/38	48.63	75	60	200	1,640	1/3, 1/3, 1/3	60	2,035	98.5	4.7	10	+0.8
6/28/38	48.21	75	60	200	1,640	1/3, 1/3, 1/3	60	1,785	89.3	5	10.55	+0.25

* Ten per cent of the nitrogen of the food is calculated as being excreted in the feces.

1.34 Kg. in fifty days and M. D. 0.97 Kg. in sixty days. The former received 28 calories per kilogram, while the latter received 33.5 calories per kilogram. Both patients were on subcaloric diets as judged by the Aub-Du Bois [8] standards.

Neither patient had nocturia. F. H. had her menstrual period at the expected time; M. D. had a period for the first time in months while living on this regimen.

Urine.—F. H. showed sugar in the urine except for seven times, and *most fractional specimens gave a 4 plus reaction.* M. D.'s urine was never sugar free during her entire stay in the hospital and *all* fractional specimens revealed glycosuria (4 plus); *no ketone bodies were present.* The volume of urine ranged between 850 and 2,500 cc. except on one occasion when M. D. had severe glycosuria with a volume of 3,700 cc. for the twenty-four hour period. The excretion of sugar was totally irregular, and, even though every attempt was made to keep the conditions of the experiment constant, it was impossible to predict whether the total dextrose excreted would be high or low. When the total excreted dextrose was subtracted from the total intake (exclusive of the dextrose derived from the protein and fat) the amounts utilized were not markedly different whether the diet was divided into ⅓, ⅓, ⅓ or ⅕, ⅖, ⅖ or any combination indicated in the tables. During the first thirty days, while receiving 200 Gm. of carbohydrate daily, or a total of 6,000 Gm., each patient excreted a total of about 3,000 Gm. of sugar—an average of 100 Gm. per day. When the carbohydate intake was reduced to 100 Gm. a day the excretion of dextrose fell definitely, but even on this regimen the division of the diet into ⅓, ⅓, ⅓ or ⅕, ⅖, ⅖ made little difference, except that the glycosuria was heavier when the diet was unevenly partitioned. Actually, F. H. excreted 128 Gm. of dextrose in twenty days with a 2,000 Gm. intake, and M. D. excreted 1,443 Gm. with a 2,000 Gm. intake.

The nitrogen balance of both patients was positive during the experimental period, though there were occasional days when they excreted more nitrogen than they ingested.

Blood Sugar Curve.—A review of the data in table 3 shows that though the sugar content of the specimens of the blood taken during fasting was variable, nevertheless it was within or close to normal limits. It is clear also that the dextrose content of all the postprandial specimens was considerably elevated and that this hyperglycemia was sustained irrespective of the type of diet used or the manner in which

8. Aub, J. C., and Du Bois, E. F.: Clinical Calorimetry: XIX. The Basal Metabolism of Old Men, Arch. Int. Med. **19**:823 (May) 1917.

it was fractionated. In the case of F. H. the blood sugar curves were lower after the first ten day period, but, as can be seen, neither the type nor the division of the diet influenced them materially. This was true in general in the case of M. D., also with perhaps a more pronounced elevation when she was receiving the higher carbohydrate diet. The division of the diet into unequal portions failed to affect either the character or the absolute values significantly.

In the record of M. D. (table 2) there is an interruption of the daily sequence from June 13 to June 20, 1938. During this period, because of a misunderstanding, she consumed a package of diabetic ginger snaps. These had little carbohydrate value but were rich in protein and fat. All the data collected during this period were therefore discarded.

This unpleasant incident caused a thorough investigation as to the reliability of data on the dietary intake. F. H. had no visitors during

TABLE 3.—*Blood Sugar Content for Patients F. H. and M. D.**

Date	8 a. m. Fasting	11 a. m.	1 p. m.	4 p. m.	7 p. m.	Diet (Gm.)	Dietary Divisions
				Patient F. H.			
3/ 7/38	192	272	333	288	328	Protein 75, fat 60, carbohydrate 200	1/3, 1/3, 1/3
3/18/38	95	180	166	205	230	Protein 75, fat 60, carbohydrate 200	1/5, 2/5, 2/5
3/31/38	64	241	226	194	220	Protein 75, fat 60, carbohydrate 200	1/3, 1/3, 1/3
4/ 8/38	55	192	214	211	276	Protein 75, fat 105, carbohydrate 100	1/3, 1/3, 1/3
4/19/38	100	185	211	220	241	Protein 75, fat 105, carbohydrate 100	2/5, 1/5, 2/5
				Patient M. D.			
5/ 2/38	150	454	428	375	375	Protein 75, fat 60, carbohydrate 200	1/3, 1/3, 1/3
5/11/38	79	365	444	357	468	Protein 75, fat 60, carbohydrate 200	1/5, 2/5, 2/5
5/21/38	172	333	340	319	375	Protein 75, fat 60, carbohydrate 200	1/3, 1/3, 1/3
5/31/38	133	306	326	365	333	Protein 75, fat 105, carbohydrate 100	1/3, 1/3, 1/3
6/10/38	68	214	272	267	340	Protein 75, fat 105, carbohydrate 100	1/5, 2/5, 2/5

* The values for the blood sugar are expressed in milligrams per hundred cubic centimeters.

her stay in the hospital and she stated most emphatically that she committed no dietary mistakes or indiscretions.

M. D. insisted that apart from eating the diabetic ginger snaps she adhered to the diet most carefully, and, after she was told that the results obtained during the dietary break would have to be discarded, she exercised most meticulous care during the last ten day period. We, therefore, are reasonably confident that no dietary breaks, other than the one mentioned, occurred. We feel that this explanation is essential, as our results are so striking as to suggest that additional food might have been ingested. To the best of our knowledge and that of our dietetic staff such was not the case with the one exception already mentioned.

COMMENT

The treatment of diabetes mellitus has been constantly changing, chiefly because of the introduction of insulin and later of protamine insulin. The attitude toward the blood sugar levels and the sugar-

free urine has changed. It may still be desirable to balance the diet with insulin so as to maintain the urine free from sugar, but today glycosuria and consequently persistent or intermittent hyperglycemia may be acceptable as adequate regulation. Then, too, the diet has been changing. There are the high carbohydrate and the low carbohydrate diet, there is the "free diet," or no diet at all, of Stolte,[9] and since the introduction of protamine insulin there has been discussion of the merits and advantages of equal and unequal divisions of the diet. Among our ambulatory patients receiving protamine zinc insulin we have observed constant and persistent glycosuria for about one and one half years. They were in excellent condition. No attempt was ever made to quantitate this sugar, as we would not have been certain as to the reliability of collection. The patients whose cases have been presented also excreted large amounts of sugar daily. This phenomenon was more marked with a high than with a low carbohydrate diet. They felt perfectly well, were symptom free and did not excrete ketone bodies in the urine. Furthermore, dividing their diets into equal or unequal portions influenced the quantitative excretion of dextrose little. The blood sugar curves were also affected little by such dietary division. True, decreasing the carbohydrate fraction caused a marked fall in the urinary dextrose, but at the same time the utilization was considerably decreased. The point that we wish to emphasize is that with the use of protamine insulin even severe glycosuria is compatible with good health, maintenance of weight and nitrogen equilibrium, as well as freedom from the symptoms of diabetes and ketosis. A similar status was observed by Raihä,[10] who studied diabetic children. With two doses of regular insulin daily, at times as high as 40 units at each injection, his patients felt well, grew, developed, resisted infection and did not excrete ketone bodies, in spite of constant and marked glycosuria and a blood sugar content during fasting of 500 mg. per hundred cubic centimeters.

It is now accepted that with the employment of protamine insulin glycosuria is permissible. Qualitatively a 4 plus reaction is allowed; quantitatively no definite limit has been established. In this connection it is significant to present the view of Joslin[11] and his associates.[12] Until recently they had always insisted on sugar-free urine. Now this group have warned the physician not to expect perfection when using protamine insulin. They have expressed more liberal views than ever before and stated that they are not discouraged if heavy glycosuria is

9. Stolte, K.: Med. Klin. **27**:831, 1931.
10. Raihä, C. E.: Acta pædiat. **19**:433, 1937.
11. Joslin, E. P.: Mil. Surgeon **82**:1, 1938.
12. White, P.: South. M. J. **31**:15, 1938.

found in the fractional specimens. They pay much more attention to the total twenty-four hour excretion. Their "standard of control, based upon the twenty-four hour quantitative specimen of urine, is no longer 100 but 90 per cent." In other words, they expressed the feeling that if a patient is receiving 150 Gm. of carbohydrate and his total excretion in twenty-four hours is 15 Gm. the condition is adequately controlled. There are others [6] who ignore as much as 40 Gm. of dextrose in a twenty-four hour specimen of urine. Our patients, during the periods studied, often excreted much more than 100 Gm. daily, and they showed no ill effects either subjectively or objectively. Obviously, at present there can be no set rule as to the quantity of dextrose one is or is not permitted to excrete if the condition is to be considered well regulated. All this leads, of course, to further speculation. Whether a patient excretes 15 Gm. of sugar or more during a twenty-four hour period, he is sure to have either continuous or intermittent hyperglycemia. That is inevitable. The question therefore arises whether hyperglycemia is deleterious. As yet there is no definite reply. It has been repeatedly stated that hyperglycemia renders a diabetic patient more susceptible to infections. It has been maintained also that hyperglycemia is an etiologic factor in the atherosclerosis of the diabetic patient. Neither of these postulates has been convincingly established, and formidable objections can be raised against most hypotheses. It is important to call attention to the fact that such theories were formulated during the preinsulin era. And no doubt during that period patients with the more severe forms of diabetes were in a deplorable state of nutrition; they were desiccated and debilitated, all of which may have been conducive to increased susceptibility to infections and degenerative lesions. Today, the diabetic patient is well nourished, whether or not he reveals glycosuria and hyperglycemia. Insulin—regular and protamine—has changed the status of the diabetic patient. With improved nutrition, even in the presence of glycosuria many of our ambulatory insulin-treated patients suffered from no more frequent or more severe infections than nondiabetic persons. Considerable doubt has been cast on the effect of hyperglycemia on infections, by Mosenthal,[13] Bayne-Jones [14] and Richardson.[15] Also Marble, White and Fernwald [16] expressed the feeling that present day diabetic patients are not more susceptible to infections than normal persons. From most carefully planned and excellently presented experiments they con-

13. Mosenthal, H. O.: Hyperglycemia: Evaluation in the Treatment of Diabetes Mellitus, J. A. M. A. **105**:484 (Aug. 17) 1935.

14. Bayne-Jones, S.: Bull. New York Acad. Med. **12**:278, 1936.

15. Richardson, R.: J. Clin. Investigation **12**:1143, 1933; **14**:389, 1935.

16. Marble, A.; White, H. J., and Fernwald, A. J.: J. Clin. Investigation **17**:423, 1938.

cluded that ". . . whole blood of diabetic patients were found to possess essentially the same phagocytic, bacteriostatic and bactericidal power against selected strains of streptococci as blood from normal controls."

Evidence that hyperglycemia and glycosuria are the etiologic factors in the vascular scleroses is not convincing. The projected hypotheses have been multiple, conflicting and confusing. The clinical observations of many investigators have not yielded uniform results. One of us (E. T.) and an associate [17] have observed roentgen evidence of calcium deposits in the tibial vessels in a young diabetic patient whose diabetes was only two months old, and we have failed to find this vascular change in patients with severe diabetes of from five to twelve years' duration who revealed glycosuria most of the time. These patients were receiving insulin and were well nourished. Experimentally, MacLeod [18] could not demonstrate vascular lesions in depancreatized dogs which were maintained in a good state of nutrition by means of insulin but revealed hyperglycemia and glycosuria for over four years. Such data convince us that the same hypotheses are not applicable to diabetic patients treated with and to those treated without insulin. We believe that the administration of insulin, no matter how unsatisfactorily we imitate the biologic mechanism, offers *protection* to the diabetic patient.

The use of insulin has been instrumental in somewhat modifying the point of view concerning the deleterious effects of hyperglycemia and glycosuria, and, furthermore, since the introduction of protamine insulin even the most conservative investigators permit glycosuria; it is therefore obvious that our criteria for so-called adequate control must be revised. Stolte [9] and Raihä [10] have been guided chiefly by the clinical response and have paid no attention to glycosuria or hyperglycemia. However, they have insisted on acetone-free urine. In this country the excretion of 15 to 40 Gm. of dextrose by patients treated with protamine insulin has been accepted as adequate control. For cases managed in our outpatient department we have no quantitative data. But in the cases here presented we have observed glycosuria with a sugar content of 100 Gm. per twenty-four hours and more. We showed also that our patients had continuous hyperglycemia, except for the specimen of blood taken during fasting. There was no ketosis. Our patients presented no symptoms, and their loss of weight was not significant. From these observations we feel that the clinical features offer a more satisfactory guide to good therapy than the chemical determinations on the blood and urine. We feel that if with a speci-

17. Tolstoi, E., and Walton, M.: Unpublished data

18. MacLeod, J. J. R.: The Fuel of Life, Princeton, N. J., Princeton University Press, 1928, p. 58.

fied quantity of protamine insulin the patient is symptom free, has no ketonuria, maintains his weight and a good state of nutrition and is able to be socially useful, the desiderata of adequate control have been fulfilled, even though glycosuria and hyperglycemia are present. And since dividing the diet unequally has not altered the situation materially it is our policy to simplify the treatment as much as possible. We recommend, therefore, an adequate caloric diet and one dose of protamine insulin sufficient to maintain the patient's weight at an optimum and more or less constant level. We recommend also that the urine be free from ketone bodies, but glycosuria is not only permitted but desirable, since it offers protection from insulin reactions. Why patients on such a regimen do well is a matter for speculation. It is our thought that, provided each patient metabolizes a quantity of carbohydrate essential for his particular metabolic needs, the excess may be excreted without damaging results.

SUMMARY

Two patients with severe diabetes were carefully observed for fifty and sixty days respectively in the metabolism service. Both patients were young women. Their caloric intake was constant at 1,640 calories, and each received a single dose of protamine zinc insulin in the morning. In one case the dose was 50 and in the other 60 units. The composition of the diet was 75 Gm. of protein, 60 Gm. of fat and 200 Gm. of carbohydrate for the first thirty days, and this diet was divided into equal portions at times (⅓, ⅓, ⅓) and into unequal portions at other periods. After this thirty day period the diet was changed to 75 Gm. of protein, 105 Gm. of fat and 100 Gm. of carbohydrate. This reduced the carbohydrate by one-half, but the total calories were kept constant by increasing the fat. Equal and unequal fractionation of this diet also was employed. The dose of insulin was always kept the same, and the effects of these procedures were observed. It was noted that the patients excreted an average of 100 Gm. of dextrose daily during the first experimental thirty day period. It was observed also that in spite of this glycosuria they were remarkably free from any and all symptoms of diabetes and their loss of weight was no greater than could be accounted for by their diet, which was subcaloric for them as judged by the Aub-Du Bois standards. The nitrogen equilibrium of the patients was maintained. Their urine was free from ketone bodies. Equal or unequal division of the diets was of no consequence in relation to the glycosuria. Decreasing the carbohydrate by 100 Gm. and increasing the fat resulted in a diminution of the glycosuria, but the utilization of the dextrose also was diminished. With the lower carbohydrate diet also there was little difference in the dextrose output, whether the diet was equally or unequally divided. The sugar curves through the day revealed normal

and approximately normal values before breakfast only. During the remainder of the day there was persistent hyperglycemia irrespective of the type of diet or its division.

CONCLUSIONS

1. When protamine zinc insulin is used to treat diabetes the glycosuria is not materially influenced by dietary shifts from equal to unequal portions.

2. With protamine zinc insulin, the glycosuria was diminished by a lower carbohydrate diet, but the utilization of dextrose was greater with the higher carbohydrate diets.

3. For patients treated with protamine zinc insulin the guiding feature of satisfactory treatment should be the maintenance of weight, freedom from symptoms and absence of ketone bodies in the urine. Glycosuria is desirable, as it affords protection from reactions.

TOTAL DIFFERENTIAL AND ABSOLUTE LEUKOCYTE COUNTS AND SEDIMENTATION RATES

DETERMINED FOR HEALTHY PERSONS NINETEEN YEARS OF AGE AND OVER

EDWIN E. OSGOOD, M.D.

INEZ E. BROWNLEE, B.A.

MABLE W. OSGOOD, B.A.

DOROTHY M. ELLIS, B.A.

AND

WILLIAM COHEN, M.D.

PORTLAND, ORE.

It is the purpose of this article to supply more accurate data than have heretofore been available for total, differential and absolute leukocyte counts and sedimentation rates of healthy adults 19 years of age and over. This is one of a series of papers on hematologic standards for healthy persons from birth through adult life. The data for other age groups has been or will be reported here and elsewhere.[1]

From the Department of Medicine, University of Oregon Medical School.

1. (*a*) Osgood, E. E.: Hemoglobin, Color Index, Saturation Index and Volume Index Standards, Arch. Int. Med. **37**:685-706 (May) 1926. (*b*) Osgood, E. E., and Haskins, H. D.: Relation Between Cell Count, Cell Volume and Hemoglobin Content of Venous Blood of Normal Young Women, ibid. **39**:643-655 (May) 1927. (*c*) Osgood, E. E., and Baker, R. L.: Erythrocyte, Hemoglobin, Cell Volume, and Color, Volume and Saturation Index Standards for Normal Children of School Age, Am. J. Dis. Child. **50**:343-358 (Aug.) 1935. (*d*) Osgood, E. E.: Normal Hematologic Standards, Arch. Int. Med. **56**:849-863 (Nov.) 1935. (*e*) Osgood, E. E.; Baker, R. L.; Brownlee, I. E.; Osgood, M. W.; Ellis, D. M., and Cohen, W.: Total, Differential and Absolute Leukocyte Counts and Sedimentation Rates for Healthy Children Four to Seven Years of Age, Am. J. Dis. Child. **58**:61 (July) 1939; (*f*) Total, Differential and Absolute Leukocyte Counts and Sedimentation Rates for Healthy Children Eight to Fourteen Years of Age, ibid., to be published. (*g*) Osgood, E. E., and others: Total, Differential and Absolute Leukocyte Counts and Sedimentation Rates for Healthy Adolescents Fifteen to Eighteen Years of Age, J. Lab. & Clin. Med., to be published. (*h*) Chuinard, E. G.; Osgood, E. E., and Ellis, D. M.: Total, Differential and Absolute Leukocyte Counts and Sedimentation Rates for Healthy Newborn Infants, to be published. (*i*) Chuinard, E. G.; Osgood, E. E., and Ellis, D. M.: Erythrocyte, Hemoglobin, Cell Volume, and Color, Volume and Saturation Index Standards for Healthy Newborn Infants, to be published. (*j*) Osgood, E. E., and others: Erythrocyte, Hemoglobin,

(Footnote continued on next page)

The values given in most textbooks appear to have been copied from one book to another since the very early studies. Apparently no attention has been paid to the study by Galambos [2] in 1912 or to the studies of Torday [3] and Miller,[4] who obtained similar results. Figures obtained by recent studies [5] of healthy persons in various parts of the world also disagree with the textbook figures, but the differences were interpreted as probably due to climate, altitude or race. Actually, the results agree well with those of Galambos [2] and with those reported here. Results of studies of the variability of leukocyte counts [6] in which many counts

---

Cell Volume, and Color, Volume and Saturation Index Standards for Healthy Adolescent Males, Fifteen Years of Age and Over, to be published; (*k*) Erythrocyte, Hemoglobin, Cell Volume, and Color, Volume and Saturation Index Standards for Healthy Females, Fifteen years of Age and Over, to be published; (*l*) Reticulocyte Counts in Healthy Newborn Infants, to be published; (*m*) Icterus Index Determinations in Healthy Newborn Infants, to be published; (*n*) Reticulocyte Counts in Healthy Adolescent Boys and Girls, to be published.

2. Galambos, A.: Ueber das normale qualitative Blutbild, Folia haemat. **13**:153-159 (April) 1912.

3. Torday, A.: Vom normalen qualitativen Blutbild, Virchows Arch. f. path. Anat. **213**:529-536 (July) 1913.

4. Miller, S. R.: The Normal Differential Leucocyte Count, Bull. Johns Hopkins Hosp. **25**:317-322 (Oct.) 1914.

5. Stammers, A. D.: The Polymorphonuclear-Lymphocyte Ratio at an Altitude of 5,750 Feet, J. Physiol. **78**:335-338 (June 12) 1933. Peterson, R. F., and Peterson, W. G.: The Differential Count at High Altitudes, J. Lab. & Clin. Med. **20**:723-726 (April) 1935. Kennedy, W. P., and Mackay, I.: The Normal Leucocyte Picture in a Hot Climate, J. Physiol. **87**:336-344 (Sept. 8) 1936. Gänsslen, M.: Regionäre Verschiedenheiten des normalen weissen Blutbildes, Deutsche med. Wchnschr. **63**:505-507 (March 26) 1937.

6. Feinblatt, H. M.: Alimentary Leukocytosis in Eighty Normal Men: A Study in Reference to the Crise Hémoclasique of Widal, J. A. M. A. **80**:613-615 (March 3) 1923. Sabin, F. R.; Cunningham, R. S.; Doan, C. A., and Kindwall, J. A.: The Normal Rhythm of the White Blood Cells, Bull. Johns Hopkins Hosp. **37**:14-67 (July) 1925. Doan, C. A., and Zerfas, L. G.: The Rhythmic Range of the White Blood Cells in Human, Pathological Leucopenic and Leucocytic States, with a Study of Thirty-Two Human Bone Marrows, J. Exper. Med. **46**:511-539 (Sept.) 1927. Shaw, A. F. B.: The Diurnal Tides of the Leucocytes of Man, J. Path. & Bact. **30**:1-19 (Jan.) 1927. Smith, C., and McDowell, A. M.: Normal Rhythm of White Blood Cells in Women, Arch. Int. Med. **43**:68-84 (Jan.) 1929. Medlar, E. M.: The Extent of Variations in the Leukocytes of Normal Individuals, Am. J. M. Sc. **177**:72-87 (Jan.) 1929. Martin, H. E.: Physiologic Leucocytosis: The Variation in the Leucocyte Count During Rest and Exercise, and After the Hypodermic Injection of Adrenaline, J. Physiol. **75**:113-129 (June 29) 1932. Lawrence, J. S.; Stephens, D. J., and Jones, E.: Studies in the Normal Human White Blood Cell Picture: II. The Effect of Digestion on the White Blood Cells, Am. J. Physiol. **106**:309-313 (Nov.) 1933. Kennon, B. R., III; Shipp, M. E., and Hetherington, D. C.: A Study of the White Blood Cell Picture in Six Young Men, ibid. **118**:690-696 (April) 1937.

were made for a few subjects also agree better with our results and those of Galambos than with the textbook figures. Garrey and Bryan [7] recently reviewed the literature on total leukocyte counts. Bryan, Chastain and Garrey [8] showed that there is a lower and less variable leukocyte count under basal conditions, but Schweizer [9] was unable to confirm these findings, and it is hardly practicable to do leukocyte counts only under basal conditions in clinical practice.

In many papers on the leukocyte count in disease it is concluded that lymphocytosis or leukopenia occurs because the counts in the disease studied differ from the textbook figures, although they are well within the normal limits here defined.

SUBJECTS

All of the subjects lived in or near Portland, Ore., at an elevation of less than 500 feet. All were white and nativeborn, in most cases of nativeborn parents. The women were chiefly nurses from the staff of Multnomah County Hospital and technicians from the laboratory of the University of Oregon Medical School. The men were mostly medical students, with a few interns and physicians. All felt well at the time the blood was drawn and had had a recent physical examination. All persons over 19 years of age were grouped together because the scatter diagrams[1d] showed no significant differences for the two sexes and no marked variation with age in this group. Few persons over 30 years of age were studied, so that these results can be strictly applied only to persons 19 to 30 years of age. It seems probable, however, that the values will prove relatively constant throughout adult life.

METHODS

Since the methods used have been described in detail elsewhere,[10] they will be only commented on here. Venous blood mixed with 2 mg. of dry potassium oxalate per cubic centimeter was used for all determinations; this method is recommended because it is more convenient and accurate than use of blood from the finger or ear. The blood was taken at any time of day, as in ordinary office practice.

The leukocyte counts were made by skilled technicians using Levy-Neubauer counting chambers and Thoma pipets. An area of 4 sq. mm. was counted, with all the precautions outlined by Osgood.[11] The differential cell counts were made on Wright's stained smears on glass slides. Buffer phosphate was used as a diluent. The details of smearing and of staining are described elsewhere.[12]

---

7. Garrey, W. E., and Bryan, W. R.: Variations in White Blood Cell Counts, Physiol. Rev. **15**:597-638 (Oct.) 1935.

8. Bryan, W. R.; Chastain, L. L., and Garrey, W. E.: The Leucocyte Count of Young Male Adults Observed After a Period of Rest and During Mild Activity in the Early Morning, Am. J. Physiol. **113**:430-440 (Oct.) 1935.

9. Schweizer, M.: The "Basal Level" of the White Cell Count in Man, Am. J. Physiol. **105**:217-219 (July 1) 1933.

10. Osgood, E. E.: A Textbook of Laboratory Diagnosis, ed. 2, Philadelphia, P. Blakiston's Son & Co., 1935, pp. 409-415 and 430-432.

11. Osgood,[10] p. 409.

12. Osgood, E. E., and Ashworth, C. M.: Atlas of Hematology, San Francisco, J. W. Stacey, Inc., 1937, p. 8.

TABLE 1.—*Total, Differential and Absolute Leukocyte Counts and Sedimentation Rates of Adults Nineteen Years Old* *

	Leukocytes per Cu. Mm.	Neutrophil Lobocytes %	Neutrophil Lobocytes Per Cu. Mm.	Neutrophil Rhabdocytes %	Neutrophil Rhabdocytes Per Cu. Mm.	Eosinophil Lobocytes %	Eosinophil Lobocytes Per Cu. Mm.	Basophil Lobocytes %	Basophil Lobocytes Per Cu. Mm.	Lymphocytes %	Lymphocytes Per Cu. Mm.	Monocytes %	Monocytes Per Cu. Mm.	Disintegrated Cells %	Disintegrated Cells Per Cu. Mm.	Sedimentation Rate 15 Min.	Sedimentation Rate 45 Min.
Men																	
	5.6	48	2.7	1	0.06	2	0.11	1	0.06	40	2.2	4	0.22	4	0.22	1.0	3.5
	6.1	57	3.5	0	0	3	0.18	0	0	37	2.3	3	0.18	0	0	0	0
	6.1	46	2.8	0	0	1	0.06	1	0.06	46	2.8	4	0.24	2	0.12	1.5	2.0
	6.4	50	3.2	0	0	1	0.06	0	0	40	2.6	9	0.58	0	0	1.0	2.0
	6.6	51	3.4	0	0	1	0.07	0	0	45	3.0	3	0.20	0	0	0	0
	6.6	40	2.6	1	0.07	1	0.07	1	0.07	48	3.2	7	0.46	2	0.13	0.5	1.0
	6.7	47	3.1	2	0.13	2	0.13	1	0.07	39	2.6	1	0.07	8	0.54	0.5	2.0
	6.7	51	3.4	1	0.07	3	0.20	0	0	37	2.5	6	0.40	2	0.13	0.5	3.0
	6.8	66	4.5	0	0	0	0	1	0.07	30	2.0	3	0.20	0	0	0.2	2.0
	7.0	50	3.5	1	0.07	2	0.14	0	0	43	3.0	2	0.14	2	0.14	1.5	4.5
	7.0	34	2.4	1	0.07	0	0	1	0 07	57	4.0	7	0.49	0	0	0.5	1.5
	7.8	40	3.1	0	0	1	0.08	2	0 16	51	4.0	4	0.31	2	0.16	0.5	1.0
	7.9	44	3.5	1	0.08	2	0.16	0	0	40	3.2	10	0.79	2	0.16	0.5	3.5
	8.6	44	3.8	0	0	2	0.17	0	0	49	4.2	2	0.17	3	0.26	0.5	1.0
	9.1	36	3.3	0	0	2	0.18	1	0.09	54	4.9	4	0.36	3	0.27	0.5	1.5
	10.5	63	6.6	0	0	0	0	0	0	29	3.0	8	0.84	0	0	1.0	7.0
Average	7.22	47.9	3.46	0.5	0.03	1.4	0.10	0.6	0.04	42.8	3.09	4.8	0.35	1.9	0.13	0.6	2.2
Maximum	10.5	66	6.6	2	0.13	3	0.20	2	0.16	57	4.9	10	0.84	8	0.54	1.5	7.0
Minimum	5.6	34	2.4	0	0	0	0	0	0	29	2.0	1	0.07	0	0	0	0
Women																	
	5.0	54	2.7	1	0.05	1	0.05	0	0	42	2.1	2	0.10	0	0	1.0	3.0
	5.4	45	2.4	2	0.11	0	0	1	0.05	48	2.6	4	0.22	0	0	1.5	9.0
	6.1	52	3.2	0	0	0	0	0	0	46	2.8	2	0.12	0	0	1.0	10.0
	6.9	69	4.8	0	0	1	0.07	0	0	26	1.8	3	0.21	0	0	2.0	15.0
	7.0	51	3.6	0	0	1	0.07	0	0	48	3.4	0	0	0	0	3.0	10.0
	7.1	45	3.2	2	0.14	1	0.07	1	0.07	50	3.6	1	0.07	0	0	1.0	5.0
	7.2	64	4.6	0	0	1	0.07	0	0	31	2.2	4	0.29	0	0	0.5	20.0
	7.3	44	3.2	0	0	2	0.15	3	0.22	46	3.4	5	0.36	0	0	1.0	10.0
	7.4	35	2.6	0	0	0	0	0	0	50	4.4	6	0.44	0	0	5.0	10.0
	7.6	29	2.2	1	0.08	1	0.08	0	0	49	3.7	5	0.38	14	1.06	0	6.0
	7.9	40	3.2	1	0.08	6	0.47	0	0	42	3.3	11	0.87	0	0	2.0	7.0
	8.0	62	5.0	0	0	1	0.08	2	0.16	32	2.6	3	0.24	0	0	3.0	21.0
	8.2	63	5.2	0	0	0	0	0	0	35	2.9	2	0.16	0	0	4.0	25.0
	8.4	49	4.1	0	0	0	0	0	0	43	3.6	1	0.08	7	0.59	1.0	6.0
	8.4	66	5.5	3	0.25	2	0.17	1	0.08	22	1.8	6	0.50	0	0	2.0	15.0
	8.6	76	6.5	0	0	0	0	0	0	21	1.8	3	0.26	0	0	2.0	11.0
	11.5	75	8.6	0	0	2	0.23	0	0	14	1.6	9	1.04	0	0	1.5	8.0
Average	7.53	54.1	4.15	0.6	0.04	1.1	0.09	0.5	0.03	38.5	2.80	3.9	0.31	1.2	0.10	1.8	11.2
Maximum	11.5	76	8.6	3	0.25	6	0.47	3	0.22	59	4.4	11	1.04	14	1.06	5.0	25.0
Minimum	5.0	29	2 2	0	0	0	0	0	0	14	1.6	0	0	0	0	0	3 0
Men and Women																	
	7.22	47.9	3.46	0.5	0.03	1.4	0.10	0 6	0.04	42.8	3.09	4.8	0.35	1.9	0.13	0 6	2.2
	7.53	54	4.15	0.6	0.04	1.1	0.09	0.5	0.03	38.5	2.80	3.9	0.31	1.2	0.10	1.8	11.2
Average	7.38	51.1	3.82	0.5	0.04	1.3	0.09	0.5	0.04	40.6	2.94	4.4	0.33	1.5	0.11	1.3	6.7
Maximum	11.5	76	6.6	6	0.25	6	0.47	3	0.22	59	4.9	11	1.04	14	1.06	5.0	25.0
Minimum	5.0	29	2.2	0	0	0	0	0	0	14	1.6	0	0	0	0	0	0

* In all tables the number per cubic millimeter represents thousands. Neutrophil lobocytes are polymorphonuclears, and neutrophil rhabdocytes, staff cells.

TABLE 2.—*Total, Differential and Absolute Leukocyte Counts and Sedimentation Rates of Adults Twenty Years Old* *

* In all tables the number per cubic millimeter represents thousands. Neutrophil lobocytes are polymorphonuclears, and neutrophil rhabdocytes, staff cells.

TABLE 2.—*Total, Differential and Absolute Leukocyte Counts and Sedimentation Rates of Adults Twenty Years Old* *

	Leukocytes per Cu. Mm.	Neutrophil Lobocytes		Neutrophil Rhabdocytes		Eosinophil Lobocytes		Basophil Lobocytes		Lymphocytes		Monocytes		Disintegrated Cells		Sedimentation Rate	
		%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	15 Min.	45 Min.
									Men								
	4.5	52	2.3	0	0	0	0	0	0	44	2.0	4	0.18	0	0	?	?
	5.2	74	3.8	2	0.10	2	0.10	0	0	20	1.0	2	0.10	0	0	1.0	6.0
	5.6	47	2.6	0	0	3	0.17	0	0	42	2.4	8	0.45	0	0	0.5	5.0
	5.7	59	3.4	5	0.28	0	0	1	0.06	32	1.8	2	0.11	0	0	0.2	2.0
	6.7	69	4.6	0	0	1	0.07	0	0	24	1.6	6	0.40	0	0	0.1	1.0
	6.7	50	3.4	0	0	5	0.34	1	0.07	35	2.3	9	0.60	0	0	1.0	6.0
	6.8	54	3.7	0	0	4	0.27	0	0	34	2.3	8	0.54	0	0	1.0	1.5
	6.8	36	2.4	0	0	1	0.07	1	0.07	54	3.7	2	0.14	6	0.41	5.0	37.0
	7.0	55	3.8	0	0	2	0.14	0	0	40	2.8	3	0.21	0	0	1.0	1.5
	7.2	49	3.5	0	0	4	0.29	0	0	44	3.2	3	0.22	0	0	0.5	2.0
	7.2	61	4.4	0	0	1	0.07	0	0	33	2.4	5	0.36	0	0	?	?
	7.3	39	2.8	0	0	2	0.15	0	0	57	4.2	2	0.15	0	0	0	1.0
	7.4	69	5.1	2	0.15	3	0.22	0	0	23	1.7	3	0.22	0	0	0.5	2.0
	7.6	72	5.5	0	0	1	0.08	0	0	24	1.8	3	0.23	0	0	0.5	2.5
	7.8	58	4.5	0	0	0	0	0	0	38	3.0	4	0.31	0	0	0.5	1.0
	9.4	56	5.3	0	0	1	0.09	0	0	42	3.9	1	0.09	0	0	0.5	2.0
	10.2	51	5.2	0	0	4	0.41	0	0	38	3.9	7	0.71	0	0	1.5	9.0
Average	7.01	55.9	3.9	0.5	0.03	2	0.14	0.2	0.01	36.7	2.59	4.2	0.30	0.4	0.02	0.9	5.3
Maximum	10.2	74	5.5	5	0.28	5	0.41	1	0.07	57	4.2	9	0.71	6	0.41	5.0	37.0
Minimum	4.5	36	2.3	0	0	0	0	0	0	20	1.0	1	0.09	0	0	0	1.0
									Women								
	5.4	59	3.2	0	0	1	0.05	0	0	31	1.7	8	0.43	0	0	1.0	5.0
	5.7	56	3.2	5	0.28	3	0.17	0	0	36	2.1	0	0	0	0	4.0	35.0
	5.8	60	3.5	0	0	4	0.23	0	0	28	1.6	8	0.46	0	0	2.5	19.0
	6.2	50	3.7	0	0	9	0.56	0	0	24	1.5	8	0.49	0	0	2.0	16.0
	6.6	48	3.2	0	0	1	0.07	0	0	46	3.0	5	0.33	0	0	0.5	1.0
	6.7	37	2.5	1	0.07	0	0	2	0.13	58	3.9	2	0.13	0	0	1.0	3.0
	7.2	54	3.9	4	0.29	1	0.07	0	0	38	2.7	3	0.22	0	0	2.0	16.0
	8.6	30	2.6	0	0	8	0.69	0	0	58	5.0	4	0.34	0	0	2.0	23.0
	9.5	39	3.7	1	0.10	0	0	1	0.10	56	5.3	3	0.28	0	0	1.0	4.0
	10.2	49	5.0	1	0.10	0	0	0	0	45	4.6	4	0.41	0	0	1.0	7.0
	10.4	73	7.6	1	0.10	3	0.31	2	0.21	14	1.5	7	0.73	0	0	1.5	12.0
	10.5	73	7.7	2	0.21	4	0.42	0	0	19	2.0	1	0.10	0	0	3.0	25.0
	10.6	35	3.7	2	0.21	2	0.21	0	0	61	6.5	0	0	0	0	2.0	12.0
Average	7.95	51.7	4.12	1.3	0.10	2.8	0.21	0.4	0.03	39.5	3.18	4.1	0.30	0	0	1.8	13.7
Maximum	10.6	73	7.7	5	0.29	9	0.69	2	0.21	61	6.5	8	0.73	0	0	4.0	35.0
Minimum	5.4	30	2.5	0	0	0	0	0	0	14	1.5	0	0	0	0	0.5	1.0
									Men and Women								
	7.01	55.9	3.90	0.5	0.03	2	0.14	0.2	0.01	36.7	2.59	4.2	0.30	0.4	0.02	0.9	5.3
	7.95	51.7	4.12	1.3	0.10	2.8	0.21	0.4	0.03	39.5	3.18	4.1	0.30	0	0	1.8	13.7
Average	7.42	54.1	3.99	0.9	0.63	2.3	0.18	0.3	0.02	37.9	2.85	4.2	0.30	0.2	0.14	1.3	9.1
Maximum	10.6	74	7.7	5	0.29	9	0.69	2	0.21	61	6.5	9	0.73	6	0.41	5.0	37.0
Minimum	4.5	30	2.3	0	0	0	0	0	0	14	1.0	0	0	0	0	0.0	1.0

* See footnote, table 1.

TABLE 3—*Total, Differential and Absolute Leukocyte Counts and Sedimentation Rates of Adults Twenty-One Years Old* *

Leukocytes per Cu. Mm.	Neutrophil Lobocytes		Neutrophil Rhabdocytes		Eosinophil Lobocytes		Basophil Lobocytes		Lymphocytes		Monocytes		Disintegrated Cells		Sedimentation Rate	
	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	15 Min.	45 Min.
								Men								
4.6	68	3.1	0	0	2	0.09	1	0.05	28	1.3	1	0.05	0	0	1.0	1.0
4.9	26	1.3	1	0.05	3	0.15	1	0.05	67	3.3	2	0.10	0	0	0.5	1.0
5.2	45	2.3	3	0.16	1	0.05	2	0.10	48	2.5	1	0.05	0	0	1.0	1.5
5.2	30	1.6	2	0.10	0	0	2	0.10	65	3.4	1	0.05	0	0	0	1.0
5.4	51	2.8	0	0	2	0.11	2	0.11	43	2.3	2	0.11	0	0	0.5	2.0
5.6	49	2.7	0	0	1	0.06	1	0.06	43	2.4	6	0.34	0	0	1.0	3.0
5.6	60	3.4	0	0	2	0.11	0	0	36	2.0	2	0.11	0	0	0	1.0
5.7	62	3.5	0	0	0	0	1	0.06	35	2.0	2	0.11	0	0	1.0	5.0
5.9	33	1.9	0	0	0	0	0	0	57	3.4	2	0.12	8	0.47	1.0	1.5
6.0	57	3.4	1	0.06	5	0.30	1	0.06	29	1.7	7	0.42	0	0	0	0
6.1	42	2.6	5	0.30	1	0.06	1	0.06	39	2.4	12	0.73	0	0	1.0	9.0
6.1	56	3.4	1	0.06	3	0.18	2	0.12	25	1.5	7	0.43	6	0.37	1.0	3.0
6.2	37	2.3	0	0	2	0.12	0	0	48	3.0	0	0	11	0.68	0.5	2.0
6.2	70	4.3	0	0	2	0.12	0	0	26	1.6	2	0.12	0	0	0.5	1.0
6.4	65	4.2	0	0	0	0	0	0	29	1.9	6	0.38	0	0	0.5	2.0
6.4	33	2.1	1	0.06	6	0.38	0	0	56	3.6	4	0.26	0	0	0	1.0
6.4	46	2.9	0	0	1	0.06	0	0	43	2.8	4	0.26	6	0.38	1.0	2.0
6.4	65	4.2	1	0.06	2	0.13	0	0	31	2.0	1	0.06	0	0	1.0	4.0
6.5	53	3.4	1	0.06	1	0.06	0	0	40	2.6	5	0.32	0	0	0.1	0.5
6.6	46	3.0	0	0	3	0.20	0	0	43	2.8	1	0.07	7	0.46	1.0	1.5
6.7	48	3.2	6	0.40	1	0.07	0	0	41	2.7	4	0.27	0	0	1.0	11.0
6.8	48	3.3	2	0.14	2	0.14	0	0	42	2.9	6	0.41	0	0	0.5	1.0
6.8	60	4.1	0	0	2	0.14	0	0	35	2.4	3	0.20	0	0	0.5	2.0
7.0	42	2.9	2	0.14	3	0.21	0	0	45	3.2	8	0.56	0	0	?	?
7.1	55	3.9	0	0	1	0.07	0	0	36	2.6	8	0.57	0	0	0.5	2.5
7.2	63	4.5	0	0	1	0.07	1	0.07	31	2.2	4	0.29	0	0	1.0	5.0
7.2	47	3.4	0	0	4	0.29	1	0.07	45	3.2	3	0.22	0	0	0.5	3.0
7.4	51	3.8	0	0	2	0.15	1	0.07	39	2.9	7	0.52	0	0	?	?
7.6	46	3.5	0	0	3	0.23	0	0	50	3.8	1	0.08	0	0	1.0	1.5
7.6	66	5.0	0	0	0	0	1	0.08	25	1.9	8	0.61	0	0	1.0	2.0

	7.7	58	4.5	0	0	1	0.08	1	0.08	37	2.8	3	0.23	0	0	0.5	2.0
	7.8	61	4.8	0	0	0	0	0	0	37	2.9	2	0.16	0	0	1.0	5 0
	7.9	43	3.4	0	0	5	0.40	2	0.16	46	3.6	3	0.24	0	0	0	1.0
	8 2	60	4 9	1	0 08	0	0	1	0 08	34	2.8	4	0.33	0	0	0.5	2.0
	8.4	49	4.1	0	0	1	0.08	1	0 08	49	4.1	0	0	0	0	0.5	1.0
	8.6	52	4.5	0	0	0	0	4	0.34	40	3.4	4	0 34	0	0	0.5	1.0
	9.2	40	3.7	1	0.09	4	0.37	0	0	51	4.7	4	0.37	0	0	1.0	6.0
	10.8	62	6.7	0	0	2	0.22	0	0	30	3.2	6	0.65	0	0	?	?
	12.2	65	7 9	7	0.85	2	0 24	1	0.12	21	2.6	4	0.49	0	0	1.0	4 0
Average	6.91	51.5	3.6	0.9	0.07	1.8	0.13	0.7	0.05	40.1	2.73	3.8	0.27	1.0	0.06	0.7	2.6
Maximum	12.2	70	7.9	7	0.85	6	0.40	4	0.34	67	4.7	12	0.73	11	0.68	1.0	11.0
Minimum	4.6	26	1.3	0	0	0	0	0	0	21	1 3	0	0	0	0	0	0
								Women									
	4 9	48	2.4	8	0.39	0	0	0	0	42	2.1	2	0.10	0	0	1.5	13.0
	5.0	67	3 4	0	0	0	0	0	0	29	1.4	4	0.20	0	0	5.0	20.0
	5 2	60	3.1	5	0.26	0	0	2	0 10	33	1.7	0	0	0	0	1.5	7.0
	6 4	59	3.8	0	0	1	0.06	2	0.13	32	2.0	6	0.38	0	0	1.5	18 0
	7.4	67	5.0	2	0.15	4	0.30	0	0	17	1.3	3	0.22	7	0.52	2 0	13 0
	7.6	44	3.3	2	0.15	1	0.08	1	0 08	51	3.9	1	0.08	0	0	3.0	21.0
	8.4	56	4.7	0	0	2	0.17	0	0	27	2.3	2	0.17	13	1.09	0.5	4 0
	9.2	63	5.8	8	0.74	1	0.09	0	0	26	2.4	2	0.18	0	0	2.0	9 0
	9.2	43	4.0	0	0	3	0.28	0	0	44	4.0	10	0.92	0	0	0 5	3 0
	13.2	83	11.0	2	0.26	0	0	0	0	14	1.8	1	0.13	0	0	1.0	5.0
Average	7.65	59	4 65	2 7	0.20	1.2	0.10	0.5	0 03	31.5	2.29	3.1	0.24	2	0.16	1.8	11 3
Maximum	13.2	83	11.0	8	0.74	4	0.30	2	0.13	51	4.0	10	0.92	13	1.09	5.0	21 0
Minimum	4.9	43	2.4	0	0	0	0	0	0	14	1.3	0	0	0	0	0.5	3.0
								Men and Women									
	6.91	51.5	3.6	0.9	0.07	1.8	0.13	0.7	0.05	40.1	2.73	3.8	0.27	0.1	0.06	0.7	2.6
	7.65	59	4 65	2.7	0.20	1.2	0.10	0.5	0.03	31.5	2 29	3.1	0.24	2	0.16	1.8	11.3
Average	7.06	53.1	3.82	1.3	0.09	1.7	0.12	0.7	0.05	38.4	2 64	3.7	0.26	1.2	0.08	0.9	4.5
Maximum	13.2	83	11.0	8	0.85	6	0.40	4	0.34	67	4.7	12	0.92	13	1.09	5.0	21.0
Minimum	4.6	26	1.3	0	0	0	0	0	0	14	1.3	0	0	0	0	0	0

* See footnote, table 1.

TABLE 4.—*Total, Differential and Absolute Leukocyte Counts and Sedimentation Rates of Adults Twenty-Two Years Old* *

	Leukocytes per Cu. Mm.	Neutrophil Lobocytes %	Neutrophil Lobocytes Per Cu. Mm.	Neutrophil Rhabdocytes %	Neutrophil Rhabdocytes Per Cu. Mm.	Eosinophil Lobocytes %	Eosinophil Lobocytes Per Cu. Mm.	Basophil Lobocytes %	Basophil Lobocytes Per Cu. Mm.	Lymphocytes %	Lymphocytes Per Cu. Mm.	Monocytes %	Monocytes Per Cu. Mm.	Disintegrated Cells %	Disintegrated Cells Per Cu. Mm.	Sedimentation Rate 15 Min.	Sedimentation Rate 45 Min.
							Men										
	4.7	55	2.6	0	0	1	0.05	0	0	43	2.0	1	0.05	0	0	0.5	1.5
	5.0	36	1.8	2	0.10	3	0.15	2	0.10	55	2.8	2	0.10	0	0	0.5	1.5
	5.6	45	2.5	0	0	2	0.11	0	0	51	2.9	2	0.11	0	0	0.5	3.0
	5.7	53	3.0	1	0.06	2	0.11	0	0	36	2.1	8	0.46	0	0	1.0	3.0
	5.8	67	3.9	2	0.12	5	0.29	0	0	22	1.3	4	0.23	0	0	1.0	4.0
	6.0	41	2.5	0	0	5	0.30	2	0.12	42	2.5	3	0.18	7	0.42	0.5	2.0
	6.3	56	3.5	0	0	0	0	0	0	34	2.1	9	0.57	0	0	0.5	2.0
	6.4	60	3.8	1	0.06	0	0	0	0	34	2.2	3	0.19	0	0	1.0	4.0
	6.4	52	3.3	0	0	6	0.38	0	0	40	2.6	2	0.13	0	0	0.5	2.0
	6.5	56	3.6	0	0	2	0.13	0	0	40	2.6	2	0.13	0	0	11.0	46.0
	6.8	46	3.1	5	0.34	2	0.14	0	0	44	3.0	3	0.20	0	0	0.5	2.0
	6.8	43	2.9	0	0	3	0.20	0	0	47	3.2	7	0.48	0	0	0	3.0
	7.0	32	2.2	1	0.07	0	0	0	0	67	4.7	0	0	0	0	0	3.0
	7.0	66	4.6	0	0	0	0	1	0.07	30	2.1	3	0.21	0	0	0.5	1.5
	7.4	58	4.3	0	0	2	0.15	1	0.07	31	2.3	8	0.59	0	0	0.5	2.0
	7.4	68	5.0	0	0	1	0.07	1	0.07	24	1.8	6	0.44	0	0	0.2	2.0
	7.6	58	4.4	0	0	0	0	1	0.08	41	3.1	0	0	0	0	0.5	1.0
	7.8	47	3.7	0	0	0	0	1	0.08	43	3.4	9	0.70	0	0	0.5	2.0
	7.8	54	4.2	0	0	1	0.08	1	0.08	35	2.7	9	0.70	0	0	0.5	3.0
	7.9	57	4.5	1	0.08	1	0.08	0	0	32	2.5	2	0.16	7	0.55	13.0	16.0
	7.9	69	5.5	0	0	0	0	0	0	25	2.2	6	0.47	0	0	0.5	1.0
	8.0	60	4.8	0	0	0	0	0	0	38	3.0	2	0.16	0	0	0.5	1.0
	8.1	45	3.6	0	0	2	0.16	1	0.08	40	3.2	6	0.49	6	0.49	1.0	2.4
	8.2	64	5.2	0	0	1	0.08	0	0	33	2.7	2	0.16	0	0	5.0	23.0
	8.4	62	5.2	0	0	2	0.17	2	0.17	27	2.3	7	0.59	0	0	1.0	2.5
	8.9	44	3.9	0	0	2	0.18	0	0	44	3.9	10	0.89	0	0	0	1.0
	9.0	42	3.8	0	0	7	0.63	0	0	48	4.3	3	0.27	0	0	?	?
	9.2	48	4.4	1	0.09	1	0.09	0	0	46	4.2	4	0.37	0	0	1.0	2.0
	9.6	55	5.3	0	0	4	0.38	1	0.10	33	3.2	7	0.67	0	0	0.5	2.0
	9.7	47	4.6	0	0	1	0.10	0	0	47	4.6	5	0.48	0	0	?	?
	10.0	54	5.4	0	0	1	0.10	1	0.10	40	4.0	4	0.40	0	0	?	?
	10.4	68	7.1	1	0.10	0	0	0	0	28	2.9	3	0.31	0	0	1.0	5.0
	11.4	79	9.0	1	0.11	1	0.11	0	0	18	2.1	1	0.11	0	0	3.5	27.0
Average	7.6	54.2	4.16	0.5	0.03	1.8	0.13	0.4	0.03	38.1	2.86	4.3	0.33	0.6	0.04	1.6	5.7
Maximum	11.4	79	9.0	5	0.34	7	0.38	2	0.17	67	4.7	10	0.89	7	0.55	13.0	46.0
Minimum	4.7	32	1.8	0	0	0	0	0	0	18	1.3	0	0	0	0	0	1.0
							Women										
	4.6	61	2.8	0	0	1	0.05	0	0	29	1.3	9	0.41	0	0	9.0	29.0
	5.6	63	3.5	1	0.06	3	0.17	0	0	29	1.6	4	0.22	0	0	1.0	8.0
	6.1	51	3.1	2	0.12	0	0	1	0.06	42	2.6	4	0.24	0	0	0.5	4.0
	6.2	37	2.3	0	0	4	0.25	0	0	53	3.3	6	0.37	0	0	0.5	3.5
	6.7	44	2.3	0	0	0	0.25	0	0	52	3.5	4	0.27	0	0	1.0	6.0
	7.0	56	3.9	2	0.14	2	0.14	0	0	27	1.9	7	0.49	6	0.42	6.0	30.0
	7.0	58	4.1	2	0.14	1	0.07	0	0	30	2.1	9	0.63	0	0	1.0	3.0
	8.2	73	6.0	2	0.16	1	0.08	0	0	20	1.6	4	0.33	0	0	2.0	5.0
	11.0	59	6.5	1	0.11	1	0.11	1	0.11	30	3.3	8	0.88	0	0	3.0	18.0
Average	6.93	55.8	3.9	1.1	0.08	1.4	0.10	0.2	0.02	34.7	2.36	6.1	0.43	0.7	0.05	2.7	11.8
Maximum	11.0	73	6.5	2	0.16	4	0.25	1	0.11	53	3.5	9	0.88	6	0.42	9.0	30.0
Minimum	4.6	37	2.3	0	0	0	0	0	0	20	1.3	4	0.22	0	0	0 5	3.0
							Men and Women										
	7.6	54.2	4.16	0.5	0.03	1.8	0.13	0.4	0.03	38.1	2.86	4.3	0.33	0.6	0.04	1.6	5.7
	6.93	55.8	3.9	1.1	0.08	1.4	0.10	0.2	0.02	34.7	2.36	6.1	0.43	0.7	0.05	2.7	11.8
Average	7.45	54.5	4.1	0.6	0.04	1.7	0.12	0.4	0.03	37.4	2.75	4.7	0.35	0.6	0.04	1.8	7.1
Maximum	11.4	79	9.0	5	0.34	7	0.38	2	0.17	67	4.7	10	0.89	7	0.55	13.0	46.0
Minimum	4.6	32	1.8	0	0	0	0	0	0	18	1.3	0	0	0	0	0	1.0

TABLE 5.—*Total, Differential and Absolute Leukocyte Counts and Sedimentation Rates of Men Twenty-Three Years Old* *

	Leukocytes per Cu. Mm.	Neutrophil Lobocytes		Neutrophil Rhabdocytes		Eosinophil Lobocytes		Basophil Lobocytes		Lymphocytes		Monocytes		Disintegrated Cells		Sedimentation Rate	
		%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	15 Min.	45 Min.
	4.4	70	3.1	0	0	0	0	0	0	22	1.0	6	0.26	2	0.09	1.0	1.0
	5.0	60	3.0	1	0.05	1	0.05	1	0.05	31	1.6	6	0.30	0	0	0	1.0
	5.0	38	1.9	1	0.05	5	0.25	0	0	53	2.6	2	0.10	0	0	0.5	1.0
	5.2	62	3.2	1	0.05	1	0.05	0	0	30	1.6	6	0.31	0	0	2.0	7.0
	5.2	43	2.2	3	0.16	0	0	0	0	39	2.0	5	0.26	10	0.52	0.5	2.0
	5.3	38	2.0	3	0.16	7	0.37	2	0.11	46	2.4	4	0.21	0	0	1.0	5.0
	5.4	64	4.1	0	0	0	0	0	0	34	1.8	2	0.11	0	0	0.5	1.0
	6.2	53	3.3	0	0	0	0	0	0	46	2.9	1	0.06	0	0	0	1.5
	6.4	63	4.0	1	0.06	1	0.06	0	0	34	2.2	1	0.06	0	0	0.5	5.0
	6.8	50	3.4	2	0.14	2	0.14	0	0	40	2.7	6	0.41	0	0	1.5	6 0
	6.8	43	2.9	0	0	0	0	0	0	50	3.4	7	0.48	0	0	?	?
	6.9	27	1.9	0	0	9	0.62	1	0.07	52	3.6	2	0.14	8	0.55	1.0	2 0
	7.0	47	3.3	0	0	0	0	0	0	48	3.4	5	0.35	0	0	1.0	5.0
	7.4	67	5.0	0	0	3	0.22	0	0	24	1.8	6	0.44	0	0	0.5	1.5
	7.6	71	5.4	0	0	8	0.61	0	0	16	1.2	5	0.38	0	0	0.5	1.0
	7.9	61	4.8	1	0.08	0	0	0	0	34	2.7	4	0.32	0	0	2 0	10 0
	8.0	50	4.0	0	0	2	0.16	3	0.24	40	3.2	5	0.40	0	0	0.5	1.0
	8.2	66	5.4	0	0	3	0.25	1	0.08	27	2.2	3	0.25	0	0	0.5	1.0
	8.4	42	3.5	1	0.08	6	0.50	2	0.17	46	3.9	3	0.25	0	0	0.5	1.5
	8.6	47	4.0	0	0	4	0.34	0	0	48	4.1	1	0.09	0	0	1.0	5 0
	8.6	60	5.2	0	0	1	0.09	0	0	31	2.7	8	0.69	0	0	0.5	2.0
	9.5	55	5.2	0	0	0	0	0	0	45	4.3	0	0	0	0	1.0	2 0
	9.6	57	5.5	0	0	8	0.77	1	0.10	32	3.1	2	0.19	0	0	1.0	1.2
	11.4	63	7.2	0	0	1	0.11	0	0	32	3.6	3	0.34	0	0	1.0	1.0
	11.7	63	7.4	0	0	0	0	0	0	31	3.6	6	0.70	0	0	2.0	12.0
	12.2	47	5.7	0	0	8	0.98	0	0	40	4.9	5	0.61	0	0	0.5	1.0
Average	7.49	54.1	4.1	0.5	0.03	2.7	0.21	0.4	0.03	37.4	2.79	4.0	0.30	0.8	0.04	0 8	3.1
Maximum	12.2	71	7.4	3	0.16	9	0.98	3	0 24	53	4.9	8	0.70	10	0.55	2.0	12 0
Minimum	4.4	27	1.9	0	0	0	0	0	0	16	1.0	0	0	0	0	0	1.0

* See footnote, table 1.

TABLE 6.—*Total, Differential and Absolute Leukocyte Counts and Sedimentation Rates of Men Twenty-Four Years Old* *

	Leukocytes per Cu. Mm.	Neutrophil Lobocytes		Neutrophil Rhabdocytes		Eosinophil Lobocytes		Basophil Lobocytes		Lymphocytes		Monocytes		Disintegrated Cells		Sedimentation Rate	
		%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	15 Min.	45 Min.
	4.8	38	1.8	0	0	1	0.05	1	0.05	55	2.6	5	0.24	0	0	?	?
	5.4	49	2.6	3	0.16	1	0.05	0	0	40	2.2	6	0.32	0	0	1.0	11.0
	5.6	51	2.9	0	0	3	0.17	0	0	43	2.4	3	0.17	0	0	0.5	1.5
	6.8	52	3.5	0	0	2	0.14	0	0	41	2.8	5	0.34	0	0	?	?
	6.8	54	3.7	0	0	2	0.14	1	0.07	39	2.7	4	0.27	0	0	?	?
	7.0	66	4.6	0	0	0	0	0	0	25	1.8	8	0.56	0	0	2.0	18.0
	7.0	56	3.9	1	0.07	0	0	0	0	41	2.9	2	0.14	0	0	0.1	1.0
	7.1	48	3.4	0	0	0	0	1	0.07	48	3.4	3	0.21	0	0	0.5	1.0
	7.6	60	4.6	0	0	2	0.15	2	0.15	34	2.6	2	0.15	0	0	0.5	1.0
	7.8	56	4.4	0	0	1	0.08	0	0	40	3.1	3	0.23	0	0	0.2	1.5
	8.1	43	3.5	0	0	0	0	0	0	56	4.5	1	0.08	0	0	1.5	6.0
	8.8	72	6.3	1	0.09	2	0.18	0	0	20	1.8	5	0.44	0	0	0	1.0
	8.8	49	4.3	0	0	7	0.62	0	0	40	3.5	4	0.35	0	0	0.5	3.0
	8.9	45	4.0	0	0	1	0.09	1	0.09	51	4.5	?	0.18	0	0	?	?
	9.0	60	5.4	0	0	3	0.27	0	0	35	3.2	2	0.18	0	0	0	1.0
	10.0	47	4.7	0	0	3	0.30	2	0.20	47	4.7	1	0.10	0	0	2.0	11.5
	12.4	66	8.2	1	0.12	3	0.37	0	0	19	2.4	11	1.36	0	0	2.0	5.0
Average	7.76	53.6	4.22	0.4	0.03	1.8	0.15	0.5	0.04	39.7	3.01	3.9	0.31	0	0	0.8	4.8
Maximum	12.4	72	8.2	3	0.16	7	0.62	2	0.20	56	4.7	11	1.36	0	0	2.0	18.0
Minimum	4.8	38	1.8	0	0	0	0	0	0	19	1.8	1	0.08	0	0	0	1.0

* See footnote, table 1.

TABLE 7.—*Total, Differential and Absolute Leukocyte Counts and Sedimentation Rates of Men Twenty-Five Years Old* *

	Leukocytes per Cu. Mm.	Neutrophil Lobocytes		Neutrophil Rhabdocytes		Eosinophil Lobocytes		Basophil Lobocytes		Lymphocytes		Monocytes		Disintegrated Cells		Sedimentation Rate	
		%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	15 Min.	45 Min.
	4.4	61	2.7	0	0	10	0.44	0	0	19	0.8	10	0.44	0	0	0.5	4.0
	5.4	60	3.2	0	0	7	0.38	0	0	27	1.5	6	0.32	0	0	?	?
	5.8	50	2.9	0	0	1	0.06	0	0	44	2.6	5	0.29	0	0	1.0	7.0
	5.8	48	2.8	0	0	1	0.06	0	0	46	2.7	5	0.29	0	0	?	?
	6.1	35	2.1	0	0	3	0.18	1	0.06	58	3.5	3	0.18	0	0	0	1.5
	6.1	55	3.4	2	0.12	0	0	1	0.06	40	2.4	2	0.12	0	0	1.0	4.0
	6.3	64	4.0	0	0	2	0.13	0	0	28	1.8	6	0.38	0	0	1.0	2.0
	6.4	45	2.9	0	0	3	0.19	1	0.06	49	3.1	2	0.13	0	0	0.5	1.0
	6.4	58	3.7	1	0.06	2	0.13	0	0	36	2.3	2	0.13	0	0	2.0	13.0
	6.8	39	2.7	0	0	4	0.27	0	0	49	3.3	2	0.14	6	0.41	1.0	4.0
	7.0	61	4.3	0	0	1	0.07	0	0	31	2.2	7	0.49	0	0	1.0	2.0
	7.2	58	4.2	0	0	1	0.07	2	0.14	33	2.4	6	0.43	0	0	0.5	1.0
	7.5	69	5.2	0	0	2	0.15	0	0	22	1.6	7	0.52	0	0	0	1.0
	7.5	46	3.4	0	0	4	0.30	2	0.15	43	3.2	5	0.38	0	0	0.5	1.0
	7.6	68	5.2	0	0	4	0.30	0	0	24	1.8	4	0.30	0	0	0	1.5
	7.7	47	3.6	2	0.15	0	0	0	0	44	3.4	7	0.54	0	0	1.5	5.0
	7.9	62	4.9	0	0	4	0.32	0	0	21	1.7	12	0.95	0	0	2.0	15.0
	7.9	56	4.4	1	0.08	0	0	0	0	39	3.1	4	0.32	0	0	1.0	2.0
	8.0	61	4.9	0	0	0	0	0	0	34	2.7	5	0.40	0	0	1.0	4.5
	8.0	54	4.3	0	0	5	0.40	0	0	40	3.2	1	0.08	0	0	0.2	3.0
	8.8	79	7.0	1	0.09	0	0	0	0	17	1.5	3	0.26	0	0	0.5	3.0
	10.8	61	6.6	0	0	10	1.08	0	0	19	2.1	10	1.08	0	0	0.5	4.0
Average	7.06	56.2	4.02	0.3	0.02	2.9	0.21	0.3	0.02	34.7	2.4	5.2	0.37	0.3	0.02	0.8	4.0
Maximum	10.8	79	7.0	2	0.15	10	1.08	2	0.15	58	3.5	12	1.08	6	0.41	2.0	15.0
Minimum	4.4	35	2.1	0	0	0	0	0	0	17	0.8	1	0.08	0	0	0	1.0

* See footnote, table 1.

TABLE 8.—*Total, Differential and Absolute Leukocyte Counts and Sedimentation Rates of Men Twenty-Six to Thirty-Eight Years Old* *

	Leukocytes per Cu. Mm.	Neutrophil Lobocytes		Neutrophil Rhabdocytes		Eosinophil Lobocytes		Basophil Lobocytes		Lymphocytes		Monocytes		Disintegrated Cells		Sedimentation Rate	
		%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	15 Min.	45 Min.
	4.4	53	2.3	1	0.04	1	0.04	0	0	42	1.8	3	0.13	0	0	1.0	2.0
	4.6	22	1.0	0	0	0	0	1	0.05	68	3.1	4	0.18	7	0.32	0.5	1.0
	5.0	37	1.8	0	0	1	0.05	1	0.05	52	2.6	9	0.45	0	0	?	?
	5.4	45	2.4	0	0	0	0	0	0	47	2.5	8	0.43	0	0	1.0	1.0
	5.6	50	2.8	0	0	2	0.11	1	0.06	43	2.4	4	0.22	0	0	?	?
	5.8	55	3.2	1	0.06	2	0.12	0	0	38	2.2	4	0.23	0	0	1.0	4.0
	5.9	36	2.1	1	0.06	0	0	0	0	60	3.5	3	0.18	0	0	1.0	2.0
	6.4	46	2.9	0	0	2	0.13	0	0	46	2.9	6	0.38	0	0	?	?
	6.5	49	3.2	1	0.06	1	0.06	1	0.06	45	2.9	3	0.20	0	0	1.0	6.0
	6.8	64	4.4	0	0	0	0	1	0.07	30	2.0	5	0.34	0	0	0.5	1.0
	6.8	68	4.6	1	0.07	0	0	2	0.14	22	1.5	7	0.48	0	0	0.5	2.0
	7.3	61	4.5	3	0.22	2	0.15	0	0	33	2.4	1	0.07	0	0	1.0	7.0
	7.5	58	4.4	0	0	2	0.15	3	0.22	37	2.8	0	0	0	0	1.0	4.0
	7.6	55	4.2	0	0	4	0.30	0	0	39	3.0	2	0.15	0	0	0.5	1.5
	7.6	51	3.9	0	0	2	0.15	1	0.08	38	2.9	8	0.61	0	0	?	?
	7.7	43	3.3	0	0	3	0.23	0	0	54	4.2	0	0	0	0	0	1.5
	8.0	46	3.7	0	0	2	0.16	0	0	50	4.0	2	0.16	0	0	1.0	3.0
	8.0	47	3.8	0	0	1	0.08	0	0	40	3.2	6	0.48	6	0.48	0	2.0
	8.1	69	5.6	1	0.08	2	0.16	0	0	25	2.0	3	0.24	0	0	0.2	2.0
	8.1	63	5.1	0	0	2	0.16	0	0	31	2.5	4	0.32	0	0	?	?
	8.2	34	2.8	4	0.33	2	0.16	1	0.08	48	3.9	4	0.33	7	0.57	0.5	2.0
	8.6	44	3.8	0	0	1	0.09	0	0	52	4.5	3	0.26	0	0	?	?
	8.8	69	6.1	2	0.18	0	0	0	0	29	2.6	2	0.18	0	0	1.0	2.5
	9.6	44	4.2	4	0.38	3	0.29	1	0.10	46	4.4	2	0.19	0	0	1.0	1.0
	10.0	75	7.5	0	0	2	0.20	0	0	23	2.3	0	0	0	0	0.5	1.0
	10.0	55	5.5	0	0	2	0.20	0	0	39	3.9	4	0.40	0	0	?	?
	10.4	51	5.3	0	0	2	0.21	1	0.10	45	4.7	1	0.10	0	0	1.0	5.0
	10.8	66	7.1	0	0	1	0.11	0	0	28	3.0	3	0.32	0	0	0.5	2.0
Average	7.48	52	3.98	0.7	0.06	1.5	0.11	0.5	0.03	41.1	2.99	3.6	0.25	0.7	0.05	0.7	2.6
Maximum	10.8	75	7.5	4	0.38	4	0.30	3	0.22	68	4.7	9	0.61	7	0.57	1.0	7.0
Minimum	4.4	22	1.0	0	0	0	0	0	0	22	1.5	0	0	0	0	0	1.0

* See footnote, table 1.

TABLE 9.—*Total, Differential and Absolute Leukocyte Counts and Sedimentation Rates of Women Twenty-Three to Thirty-Six Years Old and of Men and Women Twenty-Three Years of Age and Over* *

	Leukocytes per Cu. Mm.	Neutrophil Lobocytes		Neutrophil Rhabdocytes		Eosinophil Lobocytes		Basophil Lobocytes		Lymphocytes		Monocytes		Disintegrated Cells		Sedimentation Rate	
		%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	%	Per Cu. Mm.	15 Min.	45 Min.
									Women								
	4.6	42	1.9	2	0.09	0	0	1	0.05	55	2.5	0	0	0	0	9.0	29.0
	4.8	56	2.7	0	0	0	0	0	0	41	2.0	3	0.14	0	0	5.0	25.0
	5.5	42	2.3	0	0	0	0	1	0.06	41	2.3	9	0.50	7	0.38	1.0	4.0
	5.8	59	3.4	1	0.06	2	0.12	1	0.06	30	1.7	7	0.41	0	0	0.5	4.5
	5.9	52	3.1	1	0.06	0	0	1	0.06	43	2.5	3	0.18	0	0	?	?
	6.0	72	4.3	2	0.12	2	0.12	0	0	20	1.2	4	0.24	0	0	0.5	1.0
	6.2	52	3.2	0	0	4	0.25	1	0.06	35	2.2	8	0.50	0	0	2.0	18.0
	6.2	35	2.2	1	0.06	0	0	0	0	61	3.8	3	0.19	0	0	1.0	6.0
	6.3	74	4.7	0	0	2	0.13	0	0	23	1.4	1	0.06	0	0	1.5	8.5
	6.5	51	3.3	0	0	2	0.13	1	0.06	38	2.5	8	0.52	0	0	1.0	12.0
	7.0	53	3.7	3	0.21	0	0	1	0.07	38	2.7	4	0.28	0	0	?	?
	7.1	53	3.8	1	0.07	2	0.14	0	0	37	2.6	7	0.50	0	0	2.0	8.0
	7.3	59	4.3	1	0.07	1	0.07	0	0	32	2.3	7	0.51	0	0	1.0	8.0
	7.4	67	5.0	0	0	0	0	1	0.07	26	1.9	6	0.44	0	0	1.0	5.0
	7.8	54	4.2	0	0	3	0.23	1	0.08	37	2.9	5	0.39	0	0	1.5	14.0
	8.0	62	5.0	0	0	1	0.08	0	0	32	2.6	5	0.40	0	0	?	?
	8.2	80	6.6	0	0	0	0	1	0.08	17	1.4	2	0.16	0	0	2.0	15.0
	8.8	59	5.2	0	0	1	0.09	0	0	36	3.2	4	0.35	0	0	0.5	2.0
	8.9	62	5.5	2	0.18	2	0.18	0	0	31	2.8	3	0.27	0	0	3.0	23.0
	9.0	49	4.4	1	0.09	2	0.18	3	0.27	44	4.0	3	0.27	0	0	1.0	2.0
	10.9	51	5.6	0	0	1	0.11	0	0	41	4.5	1	0.11	6	0.65	2.0	8.0
	11.8	94	9.9	0	0	0	0	0	0	6	0.7	0	0	0	0	0.5	7.0
Average	7.27	58.1	4.29	0.7	0.05	1.1	0.08	0.6	0.04	34.7	2.44	4.2	0.29	0.6	0.05	1.9	10.5
Maximum	11.8	94	9.9	3	0.21	4	0.25	3	0.27	61	4.5	9	0.52	7	0.65	2.0	29.0
Minimum	4.6	35	1.9	0	0	0	0	0	0	6	0.7	0	0	0	0	0.5	1.0
									Men and Women								
	7.44	53.9	4.07	0.5	0.04	2.2	0.17	0.4	0.03	38.3	2.8	4.2	0.30	0.5	0.03	0.8	3.5
	7.27	58.1	4.29	0.7	0.05	1.1	0.08	0.6	0.04	34.7	2.44	4.2	0.29	0.6	0.05	1.9	10.5
Average	7.4	54.7	4.11	0.5	0.04	2	0.16	0.5	0.03	37.6	2.73	4.2	0.30	0.5	0.03	1.0	4.7
Maximum	12.4	94	9.9	4	0.38	10	1.08	3	0.27	68	4.9	12	1.36	10	0.65	9.0	29.0
Minimum	4.4	22	1.0	0	0	0	0	0	0	6	0.7	0	0	0	0	0	1.0

* See footnote, table 1.

TABLE 10.—*Summary of Total, Differential and Absolute Leukocyte Counts and Sedimentation Rates of Adults Nineteen Years of Age and Over* *

Age	Cases	Leukocytes per Cu. Mm.	Neutrophil Lobocytes %	Neutrophil Lobocytes Per Cu. Mm.	Neutrophil Rhabdocytes %	Neutrophil Rhabdocytes Per Cu. Mm.	Eosinophil Lobocytes %	Eosinophil Lobocytes Per Cu. Mm.	Basophil Lobocytes %	Basophil Lobocytes Per Cu. Mm.	Lymphocytes %	Lymphocytes Per Cu. Mm.	Monocytes %	Monocytes Per Cu. Mm.	Disintegrated Cells %	Disintegrated Cells Per Cu. Mm.	Sedimentation Rate 15 Min.	Sedimentation Rate 45 Min.
Men																		
19	16	7.22	47.9	3.46	0.5	0.03	1.4	0.10	0.6	0.04	42.8	3.09	4.8	0.35	1.9	0.13	0.6	2.2
20	17	7.01	55.9	3.90	0.5	0.03	2.0	0.14	0.2	0.01	36.7	2.59	4.2	0.30	0.4	0.02	0.9†	5.3†
21	39	6.91	51.5	3.60	0.9	0.07	1.8	0.13	0.7	0.05	40.1	2.73	3.8	0.27	1.0	0.06	0.7‡	2.6‡
22	33	7.60	54.2	4.16	0.5	0.03	1.8	0.13	0.4	0.03	38.1	2.86	4.3	0.33	0.6	0.04	1.4§	5.7§
23	26	7.49	54.1	4.10	0.5	0.03	2.7	0.21	0.4	0.03	37.4	2.79	4.0	0.30	0.8	0.04	0.8‖	3.1‖
24	17	7.76	53.6	4.22	0.4	0.03	1.8	0.15	0.5	0.03	39.7	3.01	3.9	0.31	0	0	0.8¶	4.8¶
25	22	7.06	56.2	4.02	0.3	0.02	2.9	0.21	0.3	0.02	34.7	2.40	5.2	0.37	0.3	0.02	0.8#	4.6#
26-38	28	7.48	52.0	3.98	0.7	0.06	1.5	0.11	0.5	0.03	41.1	3.00	3.6	0.25	0.7	0.05	0.7**	2.6**
Average	198	7.30	53.2	3.92	0.6	0.04	2.0	0.15	0.5	0.03	38.8	2.80	4.2	0.31	0.7	0.05	0.9††	3.7††
Maximum		12.40	79.0	9.0	7.0	0.85	10.0	0.98	4	0.34	68	4.9	12	1.36	11	0.68	13.0	46.0
Minimum		4.40	22.0	1.0	0	0	0	0	0	0	16	0.8	0	0	0	0	0	0
Women																		
19	17	7.53	54.1	4.15	0.6	0.04	1.1	0.09	0.5	0.03	38.5	2.80	3.9	0.31	1.2	0.10	1.8	11.2
20	13	7.95	51.7	4.11	1.3	0.10	2.8	0.21	0.4	0.03	39.5	3.18	4.1	0.30	0	0	1.8	13.7
21	10	7.65	59.0	4.65	2.7	0.20	1.2	0.10	0.5	0.03	31.5	2.29	3.1	0.24	2.0	0.16	1.8	11.3
22	9	6.93	55.8	3.90	1.1	0.08	1.4	0.10	0.2	0.02	34.7	2.36	6.1	0.43	0.7	0.05	2.7	11.8
23-36	22	7.27	58.1	4.29	0.7	0.05	1.1	0.08	0.6	0.04	34.7	2.44	4.2	0.29	0.6	0.05	1.9	10.5
Average	71	7.47	55.8	4.22	1.1	0.08	1.5	0.11	0.5	0.03	36.0	2.63	4.2	0.31	0.8	0.06	2.0	11.6
Maximum		13.2	94	11.0	8	0.74	9	0.69	3	0.27	61	6.5	11	1.04	14	1.09	9.0	35.0
Minimum		4.6	29	1.9	0	0	0	0	0	0	6	0.7	0	0	0	0	0	1.0
Men and Women																		
	198	7.30	53.2	3.92	0.6	0.04	2.0	0.15	0.5	0.03	38.8	2.80	4.2	0.31	0.7	0.05	0.9††	3.7††
	71	7.47	55.8	4.22	1.1	0.08	1.5	0.11	0.5	0.03	36.0	2.63	4.2	0.31	0.8	0.06	2.0	11.6
Average	269	7.35	53.9	4.01	0.7	0.05	1.8	0.14	0.5	0.03	38.1	2.76	4.2	0.31	0.7	0.05	1.2‡‡	5.9‡‡
Maximum		13.2	94	11.0	8	0.85	10	0.98	4	0.34	68	6.5	12	1.36	14	1.09	13.0	46.0
Minimum		4.4	22	1.0	0	0	0	0	0	0	6	0.7	0	0	0	0	0	0

* See footnote, table 1.
† Fifteen cases.
‡ Thirty-six cases
§ Thirty cases.
‖ Twenty-five cases.
¶ Thirteen cases.
# Twenty cases.
** Twenty-one cases.
†† One hundred and seventy-six cases.
‡‡ Two hundred and forty-seven cases.

In making the differential counts, 200 or more cells were classified according to the criteria described and illustrated in the "Atlas of Hematology," [12] care being taken to study only portions of the smears so thin that the akaryocytes (erythrocytes) were not touching each other. The absolute leukocyte counts were calculated by multiplying the total leukocyte count by the percentage of the particular cell type. Plum [13] gave data from which the probable error of any of these determinations may be calculated.

The sedimentation rates were determined [14] on the same samples of oxalated blood, a Westergren pipet being used and readings being taken after fifteen and forty-five minutes. Experience with over 80,000 determinations by this method has shown it to have the advantages of the graphic methods and to be simpler and quicker.

TABLE 11.—*Statistical and Smoothed Means and Ranges for Total, Differential and Absolute Leukocyte Counts and Sedimentation Rates of Adults Nineteen Years of Age and Over*

		Statistical Mean	Probable Error Distribution	Smoothed Mean	95% Range
Leukocytes*	Per cu. mm.	7,410 ± 70	1,145 ± 23	7,400	4,000-11,000
Neutrophil lobocytes†	%	54.3 ± 0.5	7.1 ± 0.2‡	54	33-75
	Per cu. mm.	4,085 ± 66	Skew	4,000§	1,500-7,500§
Neutrophil rhabdocytes†	%	.....	.....	1*‡	0-5*‡
	Per cu. mm.	.....	.....	50	0-200*‡
Eosinophil lobocytes†	%	.....	.....	2*‡	0-6*‡
	Per cu. mm.	.....	.....	150*‡	0-400*‡
Basophil lobocytes†	%	.....	.....	0.5†	0-2†‡
	Per cu. mm.	.....	.....	50†‡	0-200†‡
Lymphocytes†	%	37.7 ± 0.5	7.6 ± 0.2	38	15-60
	Per cu. mm.	2,782 ± 39	Skew	2,750	1,000-4,500
Monocytes†	%	.....	.....	4*‡	0-9*‡
	Per cu. mm.	.....	.....	300†‡	0-800†‡
Disintegrated cells†	%	.....	.....	3*‡	0-7†‡
	Per cu. mm.	.....	.....	250*‡	0-600*‡
Sedimentation rate	15 min.	Skew	.....	2.0†‡	0.5-5.0†‡
	45 min.	Skew	.....	10.0†‡	1.0-30.0†‡

* Applies to persons 19 years of age and over.
† Applies to persons 20 years of age and over.
‡ Applies to persons 8 to 14 years of age.
§ Applies to persons 4 to 7 and 15 years of age and over.
‖ Applies to persons 15 years of age and over.
¶ Applies to persons 4 years of age and over.

RESULTS

Tables 1 to 9 summarize the individual results for the total, differential and absolute leukocyte counts and sedimentation rates for each age and sex group. On examination of these tables and the scatter diagrams [1d] it is evident that there is no significant difference with age or sex within this group, so that it is justifiable to summarize the results for the entire group, as has been done in table 10. This table gives the averages and extreme ranges. For clinical purposes, however, the

13. Plum, P.: Accuracy of Haematological Counting Methods, Acta med. Scandinav. **90**:342-364, 1936.

14. Osgood,[10] p. 430.

most useful range of normal values is not the extreme range but that which will include about plus or minus three probable errors, or 95 per cent of healthy persons. In other words, if a result in a person of this age group falls outside of this range there is less than one chance in twenty that it is normal for that person. These 95 per cent ranges are summarized in table 11.

COMMENT

The proportion of lymphocytes is higher and of neutrophil lobocytes (polymorphonuclears) lower than the textbook figures. The probable explanation for the retention of erroneous figures in the textbooks is that these figures represent fairly closely the usual counts of patients sick enough to be attending an outpatient clinic or to be hospitalized but who are not suffering from a generally recognized cause of marked leukocytosis. The dangers of using such persons as normal subjects are obvious.

In this age group the leukocyte count is lower, the neutrophil percentage is higher and the lymphocyte percentage is lower than the corresponding figures for younger persons.[1] It is of interest that the absolute number of neutrophil lobocytes (polymorphonuclears) averages 4,000 per cubic millimeter for all ages from 4 to 30 years except for the age group 8 to 14 years.

The sedimentation rates form a skew curve, with the greatest number of determinations falling in the lower ranges. It is probable that the rate of 15 mm. in forty-five minutes, which includes 80 per cent of the results, represents the strict upper limit of normal values and that the higher rates are due to mild chronic infection in the tonsils, teeth or sinuses not detectable in the routine physical examination.

SUMMARY

There are no significant age or sex differences in the total, differential or absolute leukocyte counts or in the sedimentation rates in persons 19 to 30 years of age. Table 11 gives the data which should be most useful in interpretation of each of these determinations in persons of this age group. These results agree well with those reported by Galambos in 1912 but differ from the figures given in most textbooks.

LIMITATIONS OF BIOPSY OF STERNAL MARROW

E. V. KANDEL, M.D.*
AND
G. V. LEROY, M.D.
CHICAGO

In recent years clinical hematologists have devoted considerable attention to the cytologic structure of bone marrow removed at biopsy. Published accounts of their experience vary greatly in enthusiasm and in detail. Sufficient controversy surrounds the entire subject to justify a report of our experience at the University of Chicago Clinics and in the "Medicine A" service of St. Luke's Hospital. No attempt will be made to review or summarize the entire literature, and the papers referred to are those that amplify, controvert or illustrate our own opinions.

Marrow for study has been obtained chiefly from the tibia and the sternum; the latter source was most frequently chosen. Peabody [1] and Nordenson,[2] as well as others, have shown that the functioning marrow removed from different bones tends to be similar. In fact, the prime justification for selecting any single region from which to take biopsy material is the assumption that it is representative of the hemopoietic system as a whole. Two technics for obtaining marrow are commonly used. One requires the removal of a button of bone with its underlying marrow by means of a trephine. Usually more marrow is curetted out with a spoon. The button is fixed and decalcified, and sections are cut and stained. The curetted material may be similarly treated, although usually smears or "touch" preparations are made from it. Interpretation of fixed tissue is somewhat more difficult for the hematologist, because the cells do not look quite like their counterparts in the conventional films of peripheral blood stained with Wright's stain or a related stain. On the other hand, cellular relations are maintained, and an estimate of the cellularity of the tissue is easy and more accurate. The entire

* Douglas Smith Foundation Fellow.

From the Hematology Clinic of the University of Chicago Clinics.

1. Peabody, F.: The Pathology of the Bone Marrow in Pernicious Anemia, Am. J. Path. **3**:179, 1927; A Study of the Hyperplasia of the Bone Marrow in Man, ibid. **2**:487, 1926.

2. Nordenson, N. G.: Histologic Quantitative Studies of Normal and Pathologic Bone Marrow, Hygiea **96**:193, 1934.

surgical procedure should cause no discomfort. It ought, however, to be performed under strictly aseptic conditions, as in a regular operating room.

Removal of biopsy material by means of a needle or a trocar is easier, can be done with less apparatus and need not be performed in an operating room, although an aseptic technic should be followed. Our practice is to use a shortened 14 gage spinal puncture needle with a firm finger grip and a stilet. After inducing regional anesthesia with a 2 per cent solution of procaine hydrochloride, we introduce the needle directly through the outer lamina of the manubrium or body of the sternum. We can see no advantage in entering the marrow cavity through the articulations. The material to be studied is aspirated by gentle suction with a small syringe, and as small an amount of marrow as possible is removed. We stop suction when blood appears in the syringe; the contents of the needle and its hub amount to from 0.1 to 0.2 cc. The aspirated material may be placed in an oxalated or heparinized tube, cell counts and smears being made at one's leisure. The total cell counts are so variable, depending as they do on dilution with circulating blood when larger volumes of marrow contents are aspirated, that we have stopped doing them. (Segerdahl [3] has shown that in normal persons the standard deviation of the total white cell count is from 40 to 50 per cent of the mean; hence few counts will be significant.) We make direct smears of the material on slides before it coagulates and rely on our judgment for an estimation of the cellularity. Fat droplets are easily seen, and their presence or absence influences considerably our interpretation of the state of the marrow. Our smears are stained with Wright's stain or May Grünwald-Giemsa stain.

The marrow having been acquired and stained, the difficulty of interpretation arises. It is our custom to designate the cells according to the terminology of Downey. The smears should be studied generally at first, notice being taken of the cellularity, the amount of fat, the predominant cell types and the presence of definitely abnormal cells or parasites, if any. A crude estimate may be made of the ratio of white cells to nucleated red cells. In general, this survey supplies one with the most information. After this it is our custom to perform a differential count of 500 to 1,000 cells, using several portions of several smears. Neither of these phases of the examination should be done by laboratory technicians, but if the clinician is too busy to perform a differential count, at least he should study the slide with considerable care. The chief value of the differential count is to confirm one's impression about predominating cell types and to establish the leukocyte-

3. Segerdahl, E.: Ueber Sternalpunktionen, Acta med. Scandinav., 1935, supp. 64, p. 1.

erythrocyte ratio. Comparison of the differential counts with any of several tables of normal values at once brings out the confusing fact that the normal is variable. In table 1 are listed four such standards. We have translated (correctly, we hope) differing nomenclatures into the one we adhere to. We wish to call attention particularly to the normal values listed by Segerdahl, whose work is not widely enough known. She treated her extensive data on normal values statistically, and in the second column of her figures is given the standard deviation. She demonstrated that unless the actually observed number of any cell series is greater than her mean plus three times the standard deviation or less than the mean minus three times the standard deviation it cannot be

TABLE 1.—*Normal Values According to Several Observers* *

Cell Type	Segerdahl†		Arinkin, Average	Young and Osgood, Average	Vogel, Erf and Rosenthal, Average
	Mean	Standard Deviation			
Myeloblasts	1.32	0.57	1.7	0.4	1.6
Promyelocytes	1.35	0.76	1.9	3.2	21.5
Neutrophilic myelocytes	15.0	3.76	6.35	0.8	
Eosinophilic myelocytes	1.37	0.63	0.65	...	0.7
Neutrophilic metamyelocytes	15.69	3.22	2.4	1.8	30 2
Neutrophilic stab/band forms	10.48	3.73	...	25.0	
Neutrophilic polymorphonuclears	20.86	5.47	48.0	13.3	34.0
Eosinophilic polymorphonuclears	1.44	0.77	2.3	0.4	0 9
Basophilic polymorphonuclears	0.14	0.12	0.35	0.1	0.7
Monocytes	1.67	0.62	5.7	2.1	...
Lymphocytes	16.76	4.77	11.9	10.6	8.6
Erythroblastic series (all nucleated red cells)	12.88	4.39	12.7	14.1	29.84
Histiocytes	0.03	0.07	‡	‡	0.3
Megakaryocytes	0.03	0.05	3.35	....	0.2
Plasma cells	0.39	0.35	0.6	....	...

* Segerdahl.[3] Arinkin, A.: Die intravitale Untersuchungsmethodik des Knochenmarks, Folia haemat. **38**: 233, 1929. Young, R. H., and Osgood, E. E.: Sternal Marrow Aspirated During Life: Cytology in Health and in Disease, Arch. Int. Med. **55**: 186 (Feb.) 1935. Vogel, P.; Erf, L. A., and Rosenthal, N.: Hematological Observations on Bone Marrow Obtained by Sternal Puncture, Am. J. Clin. Path. **7**: 436 and 498, 1937.

† Segerdahl's data are based on a group of 52 men.

‡ Included with the monocytes.

considered significantly abnormal. Particular attention is called to the large size of the standard deviation. Segerdahl's figures carry weight because of the large number of biopsies of normal tissue that supplied them. It may be stated that her experience with normal values is closely paralleled by our own.

The great variability of the normal, whether expressed as the standard deviation or as the range, makes it impossible to place any reliance on subtle changes in the constitution of the marrow as seen in preparations obtained by sternal puncture. In addition to the changeability expressed so impersonally by statistical methods, two other sources of variation are seen. One is the conspicuous difference in cellularity of the several slides made. In too thick preparations only the peripheral portions stain well enough for easy identification. One also frequently sees large

aggregates of marrow cells which may be discrete enough for counting. These clumps generally yield differential counts at variance with the counts obtained from more homogeneous areas. Although accurate counting of such groups is difficult, we have reason to believe that such masses are more truly representative of the actual state of the marrow, and we seek them out. The other source of variation in results is the unconscious and purely arbitrary selection of certain regions of the several slides for counting. It was curious but not surprising to find, when we reexamined our material and recounted the cells in preparing this paper, how great was the discrepancy. Frequently four differential counts were recorded, all considerably different, and although they were

TABLE 2.—*Results of Four Differential Counts of Sternal Marrow* *

Cell Type	Counter			
	E.V.K.	G.V.L.	E.V.K.	G.V.L.
Myeloblasts	1.5	0.8	0.8	1.0
Promyelocytes	3.5	0.2	0	1.0
Neutrophilic myelocytes	4.5	8.2	3.6	10.0
Neutrophilic metamyelocytes	38.0	25.8	22.5	17.7
Neutrophilic band forms	32.0	12.5	11.8	22.6
Neutrophilic polymorphonuclears	40.5	26.2	50.2	38.9
Eosinophils, all types	0	0.2	0.6	2.0
Basophils	0	0	0	0.5
Lymphocytes	1.0	0.8	0.6	0
Monocytes	0	0	0	2 3
Basophilic erythroblasts	9.0	1.0	0.4	0.5
Polychromatic erythroblasts		22.4	9.2	3.1
Orthochromatic erythroblasts				1.0

* These counts were performed by each of us on the same material. Examination of the peripheral blood of the subject on the day biopsy was performed revealed the following values: hemoglobin, 10.9 Gm.; red blood cells, 3,900,000 per cubic millimeter, and white blood cells, 22,200 per cubic millimeter. The differential count showed 89.5 per cent polymorphonuclears, 7 per cent lymphocytes and 3.5 per cent monocytes.

not mutually exclusive for diagnostic purposes, this discrepancy has caused us to look with some suspicion on reports of workers who recognize subtle diagnostic patterns in their differential counts of sternal marrow. In table 2 is presented an example of our experience in this respect. The patient had recently recovered from lobar pneumonia, but a high leukocyte count persisted for no obvious reason. The marrow was considered normal, and in several months the leukocyte count returned to normal levels.

If considered alone—that is, without the other clinical data—and frequently even with it, the results of biopsy of sternal marrow can be misleading. Brief abstracts of illustrative cases are presented, and the differential counts for the sternal marrow and the peripheral blood for the same day are listed in table 3.

REPORT OF CASES

CASE 1.—A 54 year old man had severe unexplained anemia. There were generalized enlargement of the lymph nodes and moderate hypertrophy of the liver and spleen. Two biopsies of material from the lymph nodes disclosed only simple hyperplasia. After the repair of a rectal prolapse and administration of copious doses of an iron compound the anemia gradually improved, and the patient stopped coming to the clinic.

CASE 2.—A 59 year old man was known to have had Hodgkin's disease of nineteen years' duration. The differential count of the peripheral blood was typical of this disease. Epidural infiltration of the sacral and lumbar roots had caused paraplegia. There was no other apparent skeletal involvement.

TABLE 3.—*Differential Counts of Peripheral Blood and Sternal Marrow* in Cases 1 to 5*

	Case 1		Case 2		Case 3		Case 4		Case 5	
Peripheral blood on day of biopsy										
Hemoglobin, Gm. per 100 cc.	6.0		14.0		12.4		12.6		15.1	
Red blood cells	1,890,000		4,000,000		4,670,000		3,730,000		3,540,000	
White blood cells	6,000		7,200		9,250		16,000		14,900	
Polymorphonuclears	90		88		60		32		35	
Lymphocytes	7		10		39		67		62	
Monocytes	2		2		1		1		1	
Eosinophils	1		..		..		..		2	
Basophils	..		..		..		..		..	
Sternal marrow										
Myeloblasts	0.7	0.6	1.0	0	0	0.2	3.0	2.0	0.3	0.7
Promyelocytes	1.1	2.0	2.8	1.1	0	0.9	0.4	0.7	0.6	0.4
Myelocytes	6.0	9.1	12.1	10.9	3.3	2.1	5.0	5.0	3.3	3.1
Metamyelocytes	18.3	11.0	12.0	16.7	2.3	9.0	10.2	8.1	12.1	6.6
Band forms	16.6	23.5	34.2	28.5	25.6	32.1	20.0	25.9	4.6	11.7
Polymorphonuclears	14.2	6.5	16.7	13.7	40.3	18.2	7.0	10.0	19.0	18.3
Eosinophils (all types)	1.1	2.5	4.0	4.8	5.3	7.3	0	0.7	4.0	1.8
Basophils	0.2	0.1	0	0	0	0.2	0.2	0.2	0.2	0.4
Lymphocytes	3.1	10.0	4.5	4.5	5.0	7.9	24.0	20.2	10.6	34.5
Monocytes	0	1.1	1.4	0.6	0	2.1	0	0.5	0.6	2.5
Histiocytes	0.9	3.1	0	0.4	0	0	1.0	0.5	0.3	0
Megakaryocytes	0.4	0.1	0.5	0.4	0	0.2	0	0	0	0
Basophil erythroblasts	17.1	16.5	2.0	1.4	0.3	1.3	3.2	1.6	0	1.8
Polychromatic and orthochromatic erythroblasts	15.3	10.0	18.2	17.1	17.6	14.8	26.0	25.8	28.0	16.3

* The differential counts of cells in the sternal marrow are given in duplicate to emphasize the personal variation. The counts in the left hand column in each case are by E.V.K.

CASE 3.—A 67 year old man was known, on the basis of biopsies of tissue from the lymph nodes, to have had lymphosarcoma for seven years. At the time of the sternal puncture he had copious ascites and hydrothorax. The fluid removed by paracentesis was creamy owing to the large number of small lymphocytes that it contained.

CASE 4.—A 57 year old man had suffered with chronic lymphatic leukemia for five years. There was never any suspicion of osseous involvement. The patient died, and autopsy disclosed little heteroplasia of the marrow.

CASE 5.—A 69 year old man suffered from angina pectoris. He had moderate hyperchromic macrocytic anemia, associated with achlorhydria after stimulation with histamine. The condition was unresponsive to liver therapy. The lymphocytosis and the anemia persisted until the patient ceased to attend the clinic. The condition was never explained.

The finding of apparently normal marrow in these patients may be interpreted in various ways. In cases 1 and 5 the entire disease process was obscure, and although one might expect the marrow to be abnormal, it is unjustifiable to say that the specimens studied were not representative. In case 2, in which there certainly must have been skeletal involvement, chance must have supplied us with normal-appearing marrow. In the other 2 cases it is reasonable to think that the lymphomatous process simply had not extended to the portions of the marrow studied.

Similar in some respects to the first 5 patients were 6 additional patients with definite disturbances of the hemopoietic system. The

TABLE 4.—*Differential Counts of Peripheral Blood and Sternal Marrow* in Cases 6 to 11*

	Case 6		Case 7		Case 8		Case 9		Case 10		Case 11	
Peripheral blood on day of biopsy												
Hemoglobin, Gm. per 100 cc.	13.9		14.2		14.2		14.6		7.0		7.7	
Red blood cells	3,970,000		4,560,000		4,250,000		4,400,000		2,900,000		2,030,000	
White blood cells	2,200		2,400		4,700		3,000		6,800		5,920	
Polymorphonuclears	53		70		65		57		67		22	
Lymphocytes	42		25		24		40		26		75	
Monocytes	3		4		7		3		6		3	
Eosinophils	1		1		2		..		1		..	
Basophils	..		..		1		..		..		..	
Sternal marrow							†					
Myeloblasts	3.0	2.0	1.8	0.4	0	0.8	1.4	0	1.5	2.8	1.0	0
Promyelocytes	2.5	0.8	2.0	1.9	...	1.6	3.2	1.0	1.7	0	3.0	0.4
Myelocytes	7.2	5.2	14.2	10.4	...	9.4	6.1	6.0	0.8	2.8	3.5	5.2
Metamyelocytes	8.0	7.5	8.0	9.3	...	6.5	10.0	10.0	7.4	7.6	7.5	4.3
Band forms	16.0	22.5	16.6	22.9	...	29.0	14.1	20.8	24.1	10.0	19.0	18.1
Polymorphonuclears	6.0	6.5	17.0	14.1	...	9.0	15.8	6.0	13.3	27.3	5.0	6.9
Eosinophils (all types)	3.2	7.5	0	0.9	...	2.5	4.2	1.2	1.4	3.0	7.0	5.2
Basophils	0	0.5	0	0.4	...	0.2	0.3	0	0.2	0	0	0
Lymphocytes	7.0	4.2	7.0	5.1	...	5.7	11.2	11.6	10.6	1.0	17.5	19.3
Monocytes	1.2	0.3	2.2	1.3	...	1.0	1.2	2.0	0.4	0	0	2.2
Basophilic erythrocytes	4.2	0.6	5.0	2.3	...	1.8	1.6	2.5	14.6	39.0	33.0	4.7
Polychrome and orthochrome erythrocytes	51.0	41.9	21.2	29.1	...	30.6	40.4	37.1			2.3	31.9
Megakaryocytes	0	0	0	0.2	...	0	0.3	0.9	....	....	....	....

* The differential counts for the sternal marrow are given in duplicate as in table 3.
† This biopsy was performed just one year after the one recorded to the left.

condition of 5 of these was tentatively diagnosed as Banti's syndrome. By this we mean nonspecific fibrosis of the spleen and liver associated with refractory anemia and leukopenia and with varying evidence of hypertension in the portal vein. It is strange that such a disturbance of hemopoiesis as was indicated by the peripheral blood of these patients should be associated with no significant morphologic changes in the marrow. The differential counts for the marrow and blood of this group are given in table 4.

CASE 6.—A 40 year old woman was referred to the clinic because of considerable enlargement of the liver and spleen and because of leukopenia which defied therapy. She had moderate ascites, and esophageal varicosities were seen on fluoroscopic examination. Splenectomy was attempted but could not be com-

pleted because of dense adhesions. Biopsy of tissue from the liver revealed considerable portal fibrosis. The patient remained well, although the abnormality of the blood persisted.

CASE 7.—A 28 year old woman was found to have moderate hepatosplenomegaly and leukopenia. Esophageal varices were seen by the roentgenologist. Splenectomy was recommended, but the patient declined and did not return to the clinic.

CASE 8.—A 30 year old woman consulted us because of an enlarged liver. She had only moderate leukopenia, no anemia, no esophageal varices and no demonstrable ascites. There were definitely dilated veins over the lower part of the abdomen, suggestive caput medusae. A successful splenectomy was performed. The histologic diagnosis was splenic fibrosis of the type associated with Banti's syndrome. After the operation the liver increased in size, but the patient remained well.

CASE 9.—A 49 year old woman was first seen by us because of a purpuric tendency. There was no abnormality of the coagulation mechanism as far as we could determine. The patient had a large spleen, an enlarged liver and persistent leukopenia. There was persistent occult blood in the stools from no recognizable point. During frequent roentgen examinations for the source of bleeding, extremely large esophageal varices were discovered. An exploratory operation was performed in the hope of removing the spleen as well as being able thoroughly to examine the gastrointestinal tract. For technical reasons the spleen could not be removed. After the exploratory procedure the patient was well, and, strangely, when she felt ill a small dose of roentgen therapy to the spleen cured her. The second biopsy of sternal marrow was performed a year after the first. The blood picture was essentially unchanged.

CASE 10.—A 36 year old man was seen by us after a severe gastrointestinal hemorrhage. Moderate enlargement of the spleen was the sole physical abnormality. Thorough fluoroscopic examination failed to disclose the source of the hemorrhage and revealed no esophageal varices. Surgical exploration disclosed a few large veins around the duodenum and a greatly enlarged splenic vein. It was assumed that the condition was Banti's syndrome in the early stages. After the operation there was persistent occult blood in the stools, but no more gross hemorrhages occurred and the patient felt well.

CASE 11.—A 38 year old man with severe jaundice, low grade fever, hepatosplenomegaly and malleolar ulceration, all due to syphilis, was seen by us because of persistent anemia. Eventually, with appropriate antisyphilitic treatment, he became completely well, the spleen and the liver returning to apparently normal size.

These 6 cases had several features in common, particularly splenomegaly, anemia and leukopenia. Biopsy of marrow displayed in each a rather large, but not significantly large, percentage of nucleated red cells. This may have been due in part to the tendency to hemorrhage from which all of the patients suffered more or less. It is curious that in case 11, the only one in which the cause of the pathologic process was known, the marrow was so similar to that observed in the others. The only benefit derived from these biopsies was the assurrance that the

splenomegaly and the disturbance in the blood picture were not part of a leukemic process, and we were already certain of this.

We have mentioned that the rather high percentage of nucleated red cells in the marrow of the 6 patients just described may have been due to chronic loss of blood. This may be true, but we are able to produce from our records several instances in which it has not been true of patients with severe anemia due to loss of blood or to indefinite toxic states. The counts performed on the blood and marrow of 4 such patients are contained in table 5, and brief abstracts of the case records follow:

CASE 12.—A 22 year old man underwent a radical operation on the antrum for severe chronic sinusitis. Ten days later he had a massive hemorrhage from the nose, and the hemoglobin value fell to 7.2 Gm. per hundred cubic centimeters of blood. Sternal marrow was taken for biopsy six days later, after two transfusions and oral administration of an iron compound had raised the hemoglobin content to 12.4 Gm. per hundred cubic centimeters.

CASE 13.—A 73 year old man was admitted to the hospital because of severe diarrhea, intestinal cramps and loss of weight. A leukemoid reaction was noted in the blood. He failed rapidly and died. Autopsy disclosed severe ulcerative colitis, hemorrhage from which was the apparent cause of the anemia.

CASE 14.—A 35 year old man came to us because of severe intractable anemia. There was no external bleeding, but the stools contained much occult blood. The platelets numbered 90,000. The bleeding time was two hours and the coagulation time three minutes, but the clot did not retract. On the basis of a diagnosis of idiopathic thrombocytopenic purpura splenectomy was performed. The operation was successful. The patient recovered in a spectacular manner and was still well a year later.

CASE 15.—A 58 year old woman was admitted to the ward at St. Luke's Hospital with severe anemia and leukopenia. The platelet count was 20,000. The bleeding time was prolonged; the clotting time was normal, but the clot failed to retract. The patient stated that she had had frequent epistaxis, and with case 14 in mind a diagnosis of thrombocytopenic purpura plus leukopenia was made and splenectomy was recommended. The patient refused the operation, and medical treatment with transfusions, an iron compound, ascorbic acid and yellow bone marrow was instituted. The anemia improved, but it promptly relapsed when these medicaments were stopped. Finally, two courses of roentgen therapy to the spleen were tried. The platelets increased to 350,000 and the white cells to 7,200. The permanence of the remission was not certain.

Inspection of the biopsy specimen and determination of the number of cells in the 4 preceding cases gave little evidence as to the nature of the underlying pathologic process. A diminution of platelets was noted in cases 14 and 15, but little else. As we have mentioned, for these patients biopsy served a good purpose in a clinical sense, for it suggested to us that the severe anemia did not have as a background some severe myelopathy, such as aplastic anemia or atypical leukemia. However, we

TABLE 5.—*Differential Counts of the Peripheral Blood and the Sternal Marrow* in Cases 12 to 20*

	Case 12	Case 13	Case 14	Case 15	Case 16	Case 17	Case 18	Case 19	Case 20
Peripheral blood on day of biopsy									
Hemoglobin, Gm. per 100 cc	12.4	9.6	4.3	4.2	15.6	16.8	14.0	15.4	15.0
Red blood cells	3,510,000	2,800,000	1,170,000	1,120,000	5,200,000	4,640,000	4,700,000	5,100,000	4,790,000
White blood cells	5,200	11,000	2,700	900	11,000	16,300	8,600	9,450	9,600
Polymorphonuclears	50	68	34	56	69	59	63	60	59
Lymphocytes	36	15	58	48	24	32	28	34	42
Monocytes	8	..	8	5	6	6	4	5	2
Eosinophils	4	..	1	1	..	..	5	1	0
Basophils	..	..	..	..	..	..	..	..	1
Myelocytes	..	2 (Leukemoid reaction)	..	..	..	..	..	..	..
Sternal marrow									
Myeloblasts	0 0.2	2.0 4.0	1.0 1.9	1.6	0.4	1.4	1.0	0.7	2.5
Promyelocytes	1.0 0.4	0.8 0.2	3.5 0.2	3.0	1.3	2.0	1.0	2.0	5.5
Myelocytes	6.0 5.3	3.0 3.0	11.3 11.2	12.4	9.4	14.0	15.0	11.6	5.0
Metamyelocytes	18.2 10.5	7.0 6.0	6.8 7.6	18.0	18.9	24.0	14.6	18.4	10.0
Band forms	27.8 28.9	20.3 21.0	27.7 19.5	27.0	26.3	33.6	12.8	21.0	15.0
Polymorphonuclears	14.0 15.7	37.8 40.0	2.1 4.7	10.1	11.3	6.0	13.5	14.3	25.0
All eosinophils	1.8 2.6	2.0 2.6	1.4 0.1	5.0	6.8	2.2	3.0	2.4	3.2
Basophils	0 0	0 0.3	1.0 0	0.2	0	0.2	0.5	0.7	0.7
Lymphocytes	7.4 12.1	12.0 10.0	6.0 23.9	15.0	3.2	5.4	2.0	8.1	2.2
Monocytes	2.0 2.4	1.6 2.6	2.0 0.9	5.0	0.4	1.8	1.4	3.0	1.0
Basophilic erythroblasts	4.0 5.3	0.5 3.6	3.2 0.9	0.8	1.2	0.8	1.0	2.0	2.8
Polychromatic erythroblasts	8.4 11.3	6.0 6.5	7.8 4.2	4.0	7.2	3.0	15.1	5.4	28.7
Orthochromatic erythroblasts	5.4 8.3	7.0 ...	26.7 22.9	2.0	12.3	4.2	20.1	15.4	
Histiocytes	0.6 0.2	... 0.2	....	...	...	...	....	....	....
Megakaryocytes	... ...	... ...	.... 0.9	...	0.2	...	....	....	....

* Only the differential counts of the sternal marrow in the first three cases are given in duplicate.

were reasonably sure of the diagnosis before the biopsy was performed, and the chief reason for performing it was to illuminate, if possible, the pathologic process under observation. Nevertheless, it was encouraging to have such added assurance. Other, less spectacular instances in which the negative findings at biopsy were of use to us are also included in table 5.

CASE 16.—A 16 year old boy was brought to us by his mother because he looked ill. A brother had died the year before of acute myeloid leukemia, and the mother was terrified lest a similar disease should affect the present patient. Physical examination showed normal conditions in every respect, as did hematologic investigation.

CASE 17.—A 34 year old man was studied because of persistent leukocytosis, the white blood cell count ranging from 15,000 to 20,000. The condition was unaccompanied by fever. There was slight diarrhea, for which no cause was found. Biopsy of sternal marrow eliminated a persistent suspicion that incipient leukemia was responsible for the high white cell count. Subsequently the diarrhea became more severe, and finally terminal ileitis was diagnosed. Appropriate treatment of this condition was followed by a return of the white cell count to normal.

CASE 18.—A 44 year old man presented himself to the diagnostic clinic at St. Luke's Hospital, complaining of headaches. A roentgenogram of the skull disclosed spectacular round, oval and multiloculated regions of increased radiolucency, and multiple myeloma was justifiably suspected. After all the studies were completed the patient recalled that three years earlier a similar diagnosis had been made and he had been told that he had only a few years to live. Comparison of the films showed absolutely no change, and the abnormal areas were considered to be venous lakes or developmental anomalies.

CASE 19.—A 36 year old man was referred to one of us by Dr. C. E. Shannon, of Chicago. He had a peculiar deforming lesion of the right femur, with increasing coxa vara. Roentgenographically the medullary cavity displayed coarse secondary trabeculae, evidence of bone absorption and some cystlike areas. Diagnostic guesses varied; localized osteitis fibrosa cystica, atypical Paget's disease and atypical Gaucher's disease were suggested. The normal sternal marrow was of interest, and precise chemical studies suggested a diagnosis of hyperparathyroidism.

CASE 20.—A 31 year old man who had been doing field work in archeology in Asia Minor came to the clinic because he "felt like his malaria was returning." Careful study of the sternal marrow revealed no Plasmodia, and the patient, satisfied, had no more peculiar feelings.

Although the majority of biopsies of sternal marrow reveal nothing or are at most of academic interest (as when the diagnosis is already sufficiently obvious from other clinical studies), there are occasions when the material from the marrow cavity is helpful. Such occasions occur when definitely abnormal cells are encountered. These may be evidence of metastases or of heteroplasia or there may be parasites; but the demonstration of them is significant. Frequently they cannot be identified, but the mere finding of them may illuminate an otherwise

obscure situation. There have also been a few times when we were confronted with peculiar clinical syndromes in which the differential count of the sternal marrow was definitely diagnostic. Table 6 contains the counts in several such cases, in which the biopsy was the chief factor in explaining the condition.

CASE 21.—A 38 year old Negress was seen in consultation at the Provident Hospital. She had marked enlargement of the liver and spleen and moderate enlargement of the lymph nodes. The peripheral blood was nondescript; biopsy of the lymph nodes had not been made. The marrow contained 17 per cent of "tissue cells" similar to the cells seen in small cell lymphosarcoma. The biopsy effectively eliminated the working diagnosis of aleukemic leukemia.

CASE 22.—A 58 year old man entered the clinic because of severe pain in the back. Roentgenograms disclosed a destructive lesion of the lumbar vertebrae. The peripheral blood had a definite leukemoid reaction. The sternal marrow contained clumps of basophilic erythroblasts, many of which were multinucleated and appeared neoplastic. It was felt that a diagnosis of erythroblastic multiple myeloma was tenable.

CASE 23.—A 55 year old man was admitted to the hospital with an acute illness of several weeks' duration. He suffered frequent nosebleeds, bleeding from the gums and rectum and great weakness. The spleen and liver were not enlarged, nor were the lymph nodes. Roentgen examination showed some tiny destructive lesions of the ribs. The finding of "tissue cells," similar to round cell sarcoma cells suggested a diagnosis of lymphosarcoma. The patient died promptly, and permission for autopsy was refused.

CASE 24.—A 58 year old man had had syphilis and received tremendous amounts of bismuth and mercury compounds. He entered the clinic because of weakness and pallor. He had severe anemia which resembled pernicious anemia in some respects. Because of the history of syphilis and an enlarged liver, the diagnosis was deferred pending biopsy of sternal marrow. The marrow was typical of pernicious anemia, and the subsequent successful course under liver therapy justified the diagnosis.

CASE 25.—A 21 year old Italian was referred to the clinic because he was anemic and had a peculiar yellowish color. The most striking feature of his illness was the large number of nucleated red cells in the peripheral blood. Biopsy disclosed such a predominance of these cells that despite the age of the patient a diagnosis of chronic erythroblastic anemia of the Cooley type was made. Roentgenograms of the skull later disclosed a thickened, granular diploe like that associated with Cooley's anemia. No therapeutic measure appreciably influenced the value for hemoglobin. Splenectomy was performed and the histologic appearance of the spleen confirmed the diagnosis. No special benefits except mechanical relief followed the splenectomy.

CASE 26.—A 48 year old man entered the hospital while recovering from severe pharyngitis. A low white cell count, with many mononuclears, and some enlarged lymph nodes caused his physician to fear that acute leukemia was responsible for the illness. The first biopsy of sternal material, revealing many myelocytes, seemed to confirm this view; but another one six days later gave such radically different results, as did examination of the peripheral blood, that we were

TABLE 6.—*Differential Counts of the Peripheral Blood and the Sternal Marrow* in Cases 21 to 26*

	Case 21	Case 22	Case 23	Case 24		Case 25	Case 26
Peripheral blood on day of biopsy							
Hemoglobin, Gm. per 100 cc.	9.6	13.6	6.2	6.4	6.2	8.9	14.4
Red blood cells	1,100,000	3,790,000	2,230,000	1,480,000	1,380,000	4,610,000	4,700,000
White blood cells	18,000	12,000	22,300	4,300		19,200	3,200 13,000
Reticulocytes, per cent	..	..	....	2.4	2.0	...	.....
Polymorphonuclears	35	79	18	68	..	56	8 35
Lymphocytes	60	9	81	30	..	26	50 38
Monocytes	5	6	1	2	..	16	38 10
Eosinophils	..	1	..	..	..	..	.. ..
Basophils	..	1	..	..	..	..	.. 1
Myelocytes	..	1	..	..	..	..	.. 2
Metamyelocytes	..	4	..	..	..	..	2 7
Band forms	..	2	..	..	†	134 nucleated r.b.c. : 100 w.b.c.	2 8
Sternal marrow							
Date	....	.....	....	....	....	... 1 yr. later	1/26 2/1
Myeloblasts	0	3.0 1.1	1.0 1.2	0.6	0.5	0.1 0.5	4.0 1.0
Promyelocytes	3.0	0.3 4.3	0 0.3	1.3	1.6	0.5 0	4.0 3.0
Myelocytes	2.5	7.3 8.3	2.6 1.2	15.0	6.6	4.0 2.8	37.0 12.5
Metamyelocytes	3.5	13.6 10.2	1.6 1.2	15.4	9.9	0.1 5.0	4.5 12.0
Band forms	25.0	21.0 20.6	1.0 2.3	32.4	27.5	10.1	2.0 50.0
Polymorphonuclears	19.0	18.6 6.7	5.2 5.1		13.2	7.0 16.2	1.0 15.5
All eosinophils	3.0	2.3 4.0	0 0.4	0.6	3.2	0.2 0	0.5 0
Basophils	0	0 0	0 0	0	0	0 0	0 0
Monocytes	0	0 0.4	0 0.6	0	1.0	0.2 0.2	0 1.0
Lymphocytes	8.0	5.6 3.5	4.0 3.9	1.2	4.0	3.5 0.7	6.0 3.8
Megaloblast series	....	... ...	... ...	6.8	7.3	... ...	... ...
Basophilic erythroblasts	2.5	4.3 3.7	5.2 0.5	1.7	3.8	7.5 1.0	1.5 1.0
Polychromatic and orthochromatic erythroblasts	13.5	22.2 33.9	8.4 5.5	22.1	20.1	55.0 41.0	34.5 1.0
Histiocytes	0	0 1.0	1.0 0	0.2	0	0.5	....
Megakaryocytes	0	0.6 0.2	0 0	0.6	0	0	....
Tissue cells	17.0	... ...	68.0 78.0	...	...	... .	....

* The duplicate differential counts have same significance as in the other tables.
† The patient had been receiving potent liver extract for eighteen hours.

satisfied that what we were observing was agranulocytosis in the stages of vigorous spontaneous recovery. The patient remained well.

That biopsy of sternal marrow is useful as a scientific tool in the study of pernicious anemia is illustrated by two examples from our series of such patients who had biopsies before and shortly after receiving liver extracts. In case 24 (table 6), the potent extract was given eighteen hours before the second biopsy and no effect was apparent. However, the patient subsequently had a satisfactory reticulocyte response and a good hematologic recovery. The patient in case 27 (table 7) had received in the four days preceding the biopsy one and one-half vials of a potent liver extract and an indefinite number of

TABLE 7.—*Differential Counts of Peripheral Blood and Sternal Marrow in Cases 27 to 28*

	Case 27		Case 28			
Biopsy number	1	2	1	2	3	4
Date	2/25	3/9	10/1	10/5	10/10	10/23
Peripheral blood on day of biopsy						
Hemoglobin, Gm. per 100 cc.	6.0	7.5	6.8	6.9	6.9	8.0
Red blood cells	1,580,000	1,810,000	1,900,000	1,900,000	2,100,000	2,890,000
White blood cells	5,500	.....	4,000	.....	.....	.....
Reticulocytes, per cent	56	2	0.2	0.5	0.7	8.0
Sternal marrow						
Myeloblasts	0.4	0	1.2	1.1	1.0	0.6
Promyelocytes	1.0	0.8	3.1	3.0	2.1	1.5
Myelocytes	7.0	14.9	12.7	10.0	11.5	4.4
Metamyelocytes	8.0	14.2	6.4	4.5	7.0	8.7
Band forms	15.2	22.4	15.9	9.8	8.4	18.9
Polymorphonuclears	9.8	16.4	6.4	9.0	3.1	27.5
Lymphocytes	5.5	1.6	3.6	5.8	7.7	13.3
Basophils	...	...	...	...	...	...
Megaloblasts			12.3	15.0	3.5	0
Polychromatic and orthochromatic megaloblasts	0.8	1.6				
	...	...	12.3	19.2	7.7	0
Basophilic erythroblasts	5.5	3.2	6.9	5.0	3.5	2.6
Polychromatic erythroblasts	20.2	8.2	6.4	4.6	30.7	9.8
Orthochromatic erythroblasts	21.4	6.0	6.5	5.0	3.8	6.4

capsules of extralin (a liver and stomach preparation). The reticulocytes were 56 per cent at the time of the first biopsy, and the marrow picture was that of a typical response to liver therapy. Case 28 (table 7) is an ideal illustration of this use of biopsy. Tissue for the first biopsy was taken before any medication had been given. The second biopsy was made after four days' trial of an experimental, supposedly antianemic product. In the five days after the second biopsy there was no evidence of reticulocytosis, a fact that was predicted on the basis of the unchanged megaloblastic picture after four days of treatment. Tissue for the third biopsy was taken fourteen and one-half hours after a potent extract derived from 300 Gm. of liver had been given. The rise in reticulocytes provoked by this injection did not appear until the third day. The last biopsy was made thirteen days after the third one and revealed a complete transformation of the marrow to normal.

COMMENT

We have had unfortunately few cases in which the presence of abnormal cells in the material obtained by sternal puncture has aided us in diagnosis. Other workers [4] have cited many such instances. Obscure examples of Gaucher's disease, malaria, carcinomatosis, multiple myeloma, trypanosomiasis and kala-azar have been elucidated by biopsy of sternal marrow, and in such cases the method finds its greatest value. The separation of the leukemoid states from true leukemia and the correct diagnosis of leukemia with minimal or no changes in the peripheral blood and no clinical findings [5] is another field of usefulness for this technic. Cases 13, 22 and 26 illustrate the correct interpretation of leukemoid blood pictures. We have, strangely, seen no instances of leukemia in which the diagnosis was not established by other clinical means when the biopsy of sternal marrow was in itself diagnostic. The failure to obtain representative material is perhaps responsible for the few times when aid was anticipated and was not forthcoming.

The recognition of distinct dyscrasias of the blood solely by biopsy of sternal marrow is reported frequently.[6] These are generally aplastic

4. Rohr, K., and Hegglin, R.: Tumorzellen im Sternalpunktat, Deutsches Arch. f. klin. Med. **179**:61, 1936. Giraud, P., and Gaubert: Valeur de la ponction de la moelle osseuse pour le diagnostic du kala-azar méditerranéen, Bull. et mém. Soc. méd. d. hôp. de Paris **35**:336, 1937. Voorhoeve, H. C.: Diagnostic du paludisme au moyen de ponction sternale, Haematologica **18**:739, 1937; Case of Quartan Malaria with Nephrosis Diagnosed by Sternal Puncture, Geneesk. tijdschr. v. Nederl.-Indië **76**:3310, 1936. Fleischaker, H., and Klima, R.: Zur Diagnose des multiplen Myeloms mit Hilfe der Sternalpunktion, München. med. Wchnschr. **83**:642, 1936; Beitrag zur Kenntnis des multiplen Myeloms der plasmacellulären Leukämie und des plasmacellulären Granuloms, Folia haemat. **56**:5, 1936. Loewinger, S.: Die Bedeutung der Knochenmark- und Milzpunktion für die Diagnose des Morbus Gaucher, ibid. **53**:126, 1935. Robin, C., and Jospin, M.: Recherches expérimentales sur Trypanosoma gambiense: Présence constante du trypanosome dans la moelle osseuse; déductions pour le diagnostic et l'étude de la trypanosomiase humaine, Bull. Soc. path. exot. **30**:369, 1937.

5. Mettier, S. R., and Purviance, K.: Leukemia Without Leukocytosis (Aleukemic Myelosis) and Without Splenomegaly, Arch. Int. Med. **60**:458 (Sept.) 1937.

6. Storti, E.: Ueber ein Fall von lymphatischer Leukämie mit ausschliesslicher Lokalisation im Knochenmark und über die Bedeutung der Sternalpunktion für die Diagnose dieser Krankheitsform, Deutsches Arch. f. klin. med. **180**:612, 1937. Benhamou, E.; Nouchy, A., and Cohen-Solal, G.: Anémie aplastique précocement décelée par le médullogramme, Sang **11**:763, 1937. Fiessinger, N.; Dupuy, R., and Laur, C. M.: Les myélogrammes au cours des cirrhoses, ibid. **11**:89, 1937. Freeman, W.: Bone Marrow Studies in Glandular Fever, Am. J. Clin. Path. **6**:185, 1936. Santoianna, G.: Primi risultati di ricercha in vivo sulla del midollo osseo nel corso di alcuna dermatosi, Riforma med. **52**:1727, 1936. Kingery, L. B.; Osgood, E. E., and Illge, A. H.: Sternal Puncture: A Diagnostic Aid in Leukaemia Cutis; Possible Aid in Differentiating Lymphoblastomas, Arch. Dermat. & Syph. **35**:910 (May) 1937.

anemia and megaloblastic anemia—as in pregnancy, infectious mononucleosis or leukemia cutis. In our series there were only 3 cases (24, 25 and 26) in which the diagnosis was made in this manner, and, curiously, except in case 25 a correct diagnosis would have been made by other clinical means in a few days.

The negative value of the biopsy of sternal marrow is large, and perhaps our greatest pleasure has been in cases in which the examination allowed us to reassure patients who were thought to be seriously afflicted. This negative usefulness, however, is mitigated by the frequency with which we encountered apparently normal marrow in the face of obviously severe disease.

It is our impression and belief that extremely thorough clinical investigation of patients robs biopsy of sternal marrow of much of its value. In our hands it has more commonly been a link, and often a superfluous link, in the diagnostic chain than it has been the sole means of establishing a diagnosis. The knowledge that potent products for the treatment of pernicious anemia convert the megaloblastic marrow to erythroblastic marrow within, usually, twenty-four hours is valuable in clinical research. It permits more rapid assaying of new remedies and allows one to expose his patients to a minimum of risk during the testing period. With severely anemic patients there is an obvious advantage in not waiting seven or ten days for the expected response from experimental therapy.

SUMMARY AND CONCLUSIONS

The technics for biopsy of marrow, the normal differential counts of material so obtained and the limitations of the methods are discussed.

Our experience is reviewed, and 28 examples illustrating the uses, benefits and disadvantages of biopsy of sternal marrow are presented.

It is indicated that thorough clinical investigation deprives the biopsy of sternal marrow of many of its uses.

The usefulness of this type of biopsy in clinical research, particularly in studies of pernicious anemia, is pointed out.

METASTATIC CALCIFICATION

REPORT OF TWO CASES

DAVID M. GRAYZEL, M.D., PH.D.
AND
MAX LEDERER, M.D.
BROOKLYN

Calcinosis is the term applied to a condition marked by a widespread abnormal deposition of calcium salts in various tissues of the body. In some conditions, such as dystrophic calcification, one must assume a previous retrogressive change in the tissues with subsequent deposition of calcium salts. In other conditions, however, the lime salts are deposited in previously healthy tissues. In the latter instance there are, owing to some still unexplained factor, mobilization of the calcium from the normal depots in bones and redistribution to various tissues and organs in the body. Oversaturation of the blood with calcium salts occurs in some cases. This condition was first recognized by Virchow,[1] who called it "metastatic calcification." Since then many reports have appeared in the literature.[2] Such calcification is not limited to any one condition but has been reported in association with a variety of diseases, such as scleroderma, dermatomyositis, nephritis, renal rickets, lipoid dystrophies, leukemias and various diseases of the bone. Rothstein and Welt [3] have recently published an excellent review of the literature with a report of 3 instances of calcification in children.

From the Department of Laboratories, Division of Pathology, the Jewish Hospital of Brooklyn.

1. Virchow, R.: Ueber Kalkmetastase, Virchows Arch. f. path. Anat. **8**: 103, 1855.

2. Pusitz, M. E.; Owen, A. K., and Finney, G. A.: Calcinosis Cutis, J. A. M. A. **110**:360 (Jan. 29) 1938. Gould, S. E., and Raiford, F. T.: Calcinosis Universalis: Report of a Case with Autopsy Findings, Am. J. Roentgenol. **39**:741, 1938. Widmann, B. P.; Ostrum, N. H., and Freed, H.: Practical Aspects of Calcification in the Various Body Tissues, Radiology **30**:598, 1938. Schatz, T.: Diffuse Interstitial Calcinosis, ibid. **22**:54, 1934. De Santo, D. A.: Metastatic Calcification in Myelogenous Leukemia, Am. J. Path. **9**:105, 1933. Sheldon, J. H.: Calcinosis Universalis, Proc. Roy. Soc. Med. **27**:623, 1934. Hein, B. J.: Calcinosis Universalis, Arch. Surg. **26**:389 (March) 1933.

3. Rothstein, J. L., and Welt, S.: Calcinosis Universalis and Calcinosis Circumscripta in Infancy and Childhood: Three Cases of Calcinosis Universalis, with Review of the Literature, Am. J. Dis. Child. **52**:368 (Aug.) 1936.

The present paper is a report of 2 cases. In 1 the condition was associated with leukemic myelosis, and in the other it complicated a normal pregnancy in an otherwise apparently healthy young woman. The widespread distribution of the lesions in these 2 cases prompted this report.

REPORT OF CASES

CASE 1.—H. K., a 13 year old white girl, was admitted to the Jewish Hospital of Brooklyn on Nov. 19, 1934. She complained of weakness, pain

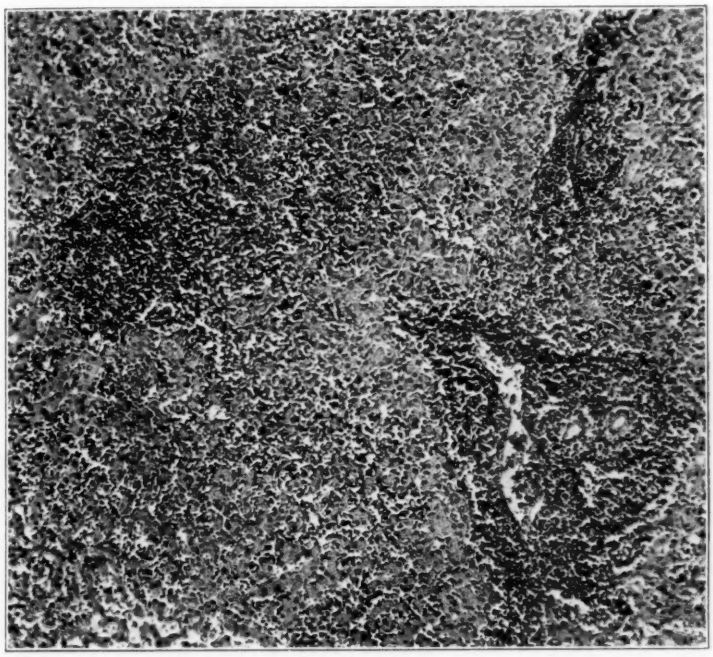


Fig. 1 (case 1).—Liver with leukemic infiltration. Hematoxylin and eosin; × 160.

in the left flank for the past twelve hours, postprandial vomiting for the past month and loss of 25 pounds (11.3 Kg.) in the last two months. The family history was irrelevant.

Two months before the present admission the patient began to have pain which started in the thighs and radiated to the ankles and feet. Later it became localized in the right flank and was more severe. She spent three weeks in another hospital, where various studies were made but no definite diagnosis was arrived at. She was discharged unimproved. The pain in the right flank disappeared, but pain developed in the left flank and shortly there-

after in the lower two ribs in the axillas. Her loss of weight was due chiefly to the fact that she was afraid to eat because of the postprandial vomiting.

The temperature on admission was 98.6 F. The pulse rate was 95, the respiratory rate 16 and the blood pressure 140 mm. of mercury systolic and 110 diastolic. The heart and lungs were normal. There was tenderness over the twelfth rib on the left side. The right kidney was palpable, and there was enlargement of the cervical, axillary, inguinal and abdominal lymph nodes. Electrocardiographic studies revealed myocardial and auricular involvement and sinus tachycardia. Roentgenographic studies of the chest, ribs, skull, hands and sella turcica showed

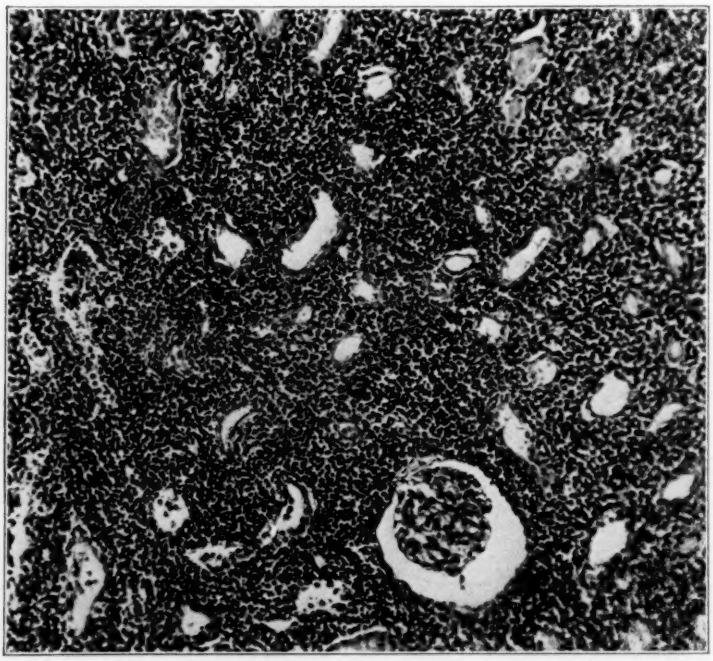


Fig. 2 (case 1).—Kidney with leukemic infiltration. Hematoxylin and eosin.

these structures to be normal. There were rarefaction and mottling in the shaft of the fibula and in the thoracic and lumbar vertebrae. There were also punched-out semicystic formations below the heads of both humeri. While the patient was in the hospital the course was progressively down hill. She had convulsions and became semicomatose. She died on February 7.

Laboratory Data.—Urinalysis revealed a fixed specific gravity of 1.008 to 1.010, a trace of albumin and a few hyaline casts but no red blood cells. The phenolsulfonphthalein test showed a total excretion of 27 per cent. A blood count showed: red cells, 3,400,000 per cubic millimeter; white cells, 6,300 per cubic millimeter, with 85 per cent polymorphonuclear neutrophilic leukocytes and 15 per cent lymphocytes. The hemoglobin content was 60 per cent.

A study of the blood chemistry on December 10 gave the following values: chlorides, 424 mg.; nonprotein nitrogen, 101 mg.; total proteins, 6.22 mg.; albumin, 3.47 mg.; globulin, 2.75 mg.; cholesterol, 197 mg.; sugar, 134 mg.; calcium, 20.6 mg., and phosphorus, 4.7 mg., per hundred cubic centimeters. The value for phosphatase was 4.5 units, the carbon dioxide content 53.5 volumes per cent and the albumin-globulin ratio 1.26. The Wassermann reaction was negative.

Clinically the condition was thought to be aleukemic myelosis.

Necropsy.—The body was well developed but poorly nourished. There was no excess fluid in the peritoneal or pleural cavities.

The heart weighed 210 Gm. The right side of the heart and the valves showed nothing of note except slight thickening at the bases of the aortic

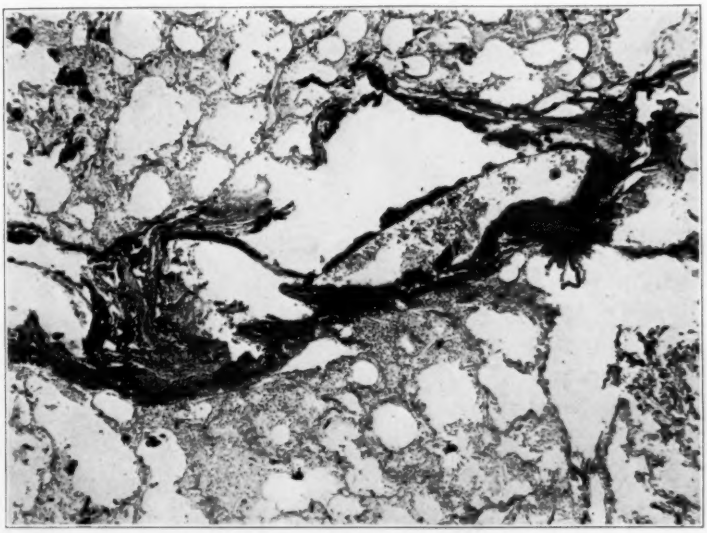


Fig. 3 (case 1).—Lung with calcification in the walls of the veins and in the parenchyma. Hematoxylin and eosin.

cusps. In the left atrium there were numerous small, sharply defined calcific plaques beneath the endocardium.

The lungs were solid; numerous irregularly shaped hard nodules could be palpated throughout. The surfaces were smooth and mottled yellow-red and purple-blue. The cut surfaces were granular; the color was a peculiar yellow with scattered areas of pink. In the areas of hardening there were gritty yellow plaques presenting the appearance of a honeycomb. The larger branches of the pulmonary artery were calcified. Histologic examination revealed calcium deposits in the walls of alveoli, in the walls of some bronchi immediately beneath the basement membrane of the lining epithelium and in the intimae of many veins and some arteries. There was no evidence of any inflammatory reaction in the calcified areas.

The spleen weighed 580 Gm. It was firm and red. In the cut surfaces the malpighian bodies were prominent. Irregular patches of yellow were distributed throughout the deep red pulp. Histologically there was widespread leukemic infiltration of the organ.

Leukemic infiltration was also noted in the liver, kidney, thymus gland, pancreas, lymph nodes and bone marrow of the ribs and vertebrae.

Minute areas of calcification were present in the adrenal glands, kidneys and thymus gland.

The parathyroid glands were small and presented nothing of note.

The anatomic diagnosis was leukemic myelosis in the bone marrow, lymph nodes, spleen, liver, kidneys, pancreas and thymus gland; metastatic calcification

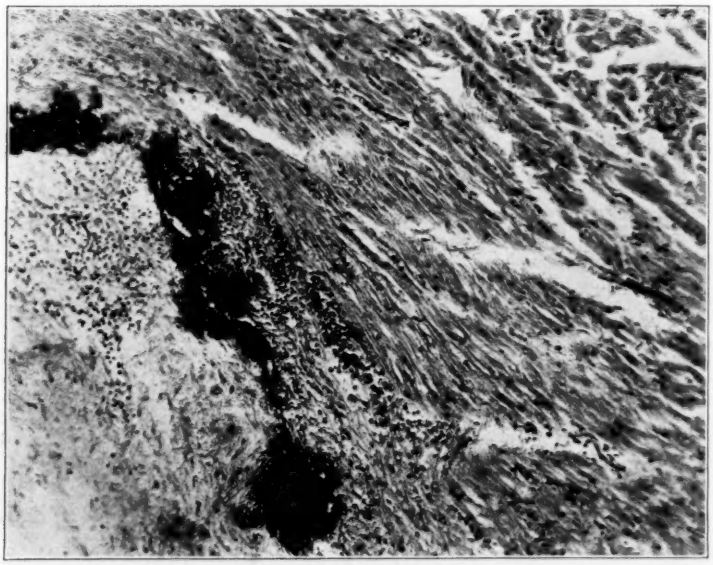


Fig. 4 (case 1).—Calcium deposits about the vessels in the wall of the left auricle. Hematoxylin and eosin; × 150.

in the lungs, left atrium, adrenal glands, kidneys, thymus gland and some of the moderate-sized arteries.

Case 2.—H. K., a 25 year old white woman, was admitted to the Jewish Hospital of Brooklyn on Oct. 17, 1937, complaining of weakness, dizziness, cough, anorexia and fever for the past six weeks. The family history was irrelevant.

The patient had had chorea at the age of 15 years. The condition lasted for three or four months. For some time afterward she had occasional attacks of involutary twitching. There was no history of cardiac involvement. She had been married for six years and had two children, a boy aged 5 years and another aged 1½ months.

About the seventh month of her second pregnancy, which terminated on September 1 in a spontaneous normal delivery, the patient began to complain of a dry, irritating cough. At home, after her discharge from the hospital, she complained of weakness, lassitude, cough and frequent attacks of vertigo. The weakness persisted, and two weeks prior to admission she consulted a physician, who found pus in her urine and treated her for "inflammation of the kidneys." The patient at no time complained of chills, pain in the back or in the lumbar regions, dysuria, hematuria or frequency of urination. She had no pains in the chest, no hemoptysis and no night sweats. Her cough was productive of a moderate amount of white sputum. On the day prior to admission her temperature rose to 104 F., and she was advised to come into the hospital.

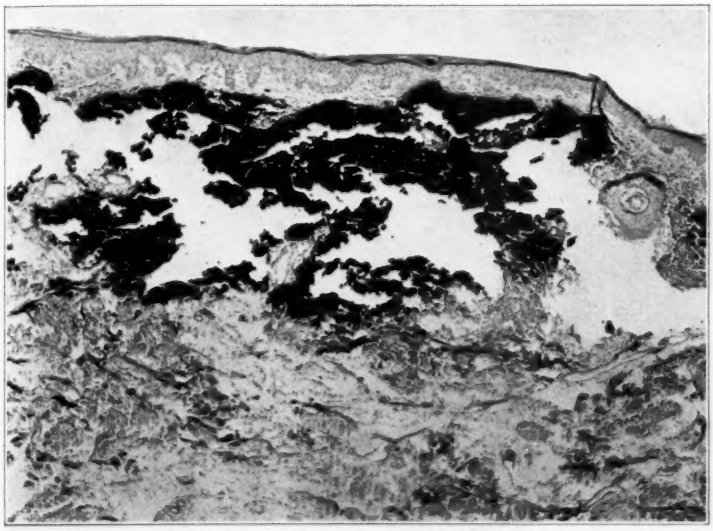


Fig. 5 (case 2).—Cutaneous lesion with extensive calcification in the upper layers of the cutis. Hematoxylin and eosin; × 45.

On her admission the temperature was 100.4 F., the pulse rate 120, the respiratory rate 20 and the blood pressure 124 mm. of mercury systolic and 60 diastolic. The heart was not enlarged. The sounds were of good quality; the beat was rapid but regular. The lungs on percussion were resonant except at the base of the right lung posteriorly; this region was dull to flat up to the angle of the scapula. Over this area the tactile fremitus was diminished. The breath sounds were vesicular but markedly diminished over the base of the right lung. There were no rales or friction rub. The liver was enlarged and moderately tender. Its edge was firm and was palpable 5 fingerbreadths below the costal margin. The spleen was not palpable. There was marked tenderness in localized areas along the tibias. There were no swellings or masses in these areas. The reflexes were hyperactive but equal.

Roentgenograms taken on the day after the patient's admission to the hospital revealed the presence of fluid at the base of the right lung. Thoracentesis was performed two days later, and 60 cc. of clear straw-colored fluid was obtained. Similar fluid was obtained on several occasions during the patient's stay in the hospital, at times as much as 1,700 cc. During this time the temperature ranged from 100 to 102 F., with an occasional rise to 104 F. On November 13 there appeared at the periphery of each breast a lesion characterized by thickening of the skin and well circumscribed smooth pearly nodules. The following day there were numerous yellow indurated nodules in both axillas, at the periphery of each breast and over the right tibia. These ranged from the size of a millet seed to that of a hazelnut. They were irregularly oval and were not tender. Some of the lesions had coalesced and become hard cardboard-like plaques. The cutaneous lesions progressed and three

Fig. 6 (case 2).—Lung with edema and calcification of the wall of the vessel and calcification of the parenchyma. Hematoxylin and eosin; × 75.

days later were present in both groins, extending along the inner aspects of both thighs. The temperature rose to 104.8 F. The patient became markedly dyspneic and cyanotic. The cardiac rate was rapid, and the sounds were of poor quality. There was dependent edema in the sacral region and in the lower extremities. Tissue from a cutaneous lesion, taken for biopsy on November 19, was described as "skin with atrophy." The course thereafter was rapidly downhill. The cutaneous lesions had advanced to involve both inguinal regions, extending upward over the iliac crests and downward along the medial and posterior aspects of the thighs to the knee of the right leg and to the midcrural region on the left leg. The patient became semistuporous on November 26. The temperature rose to 105 F.; the pulse rate rose to 150 and the pulse was weak. Fluid was present at the bases of both lungs. On the following day there was Cheyne-Stokes respiration; the cyanosis increased, and the patient died at 9:30 p. m.

Laboratory Data.—Urinalysis on October 27 showed an acid urine with a specific gravity of 1.016, a 1 plus reaction for albumin and no sugar. Numerous white blood cells were observed on microscopic examination. The blood count was as follows: hemoglobin, 53 per cent; red cells, 3,400,000 per cubic millimeter; white cells, 6,700 per cubic millimeter; polymorphonuclear leukocytes, 78 per cent; lymphocytes, 14 per cent; monocytes, 4 per cent, and band forms, 4 per cent. On November 8 the white cell count rose to 24,400 per cubic millimeter, with 64 per cent polymorphonuclear leukocytes, 12 per cent lymphocytes, 20 per cent band forms and 4 per cent monocytes. A complete study of the blood chemistry gave the

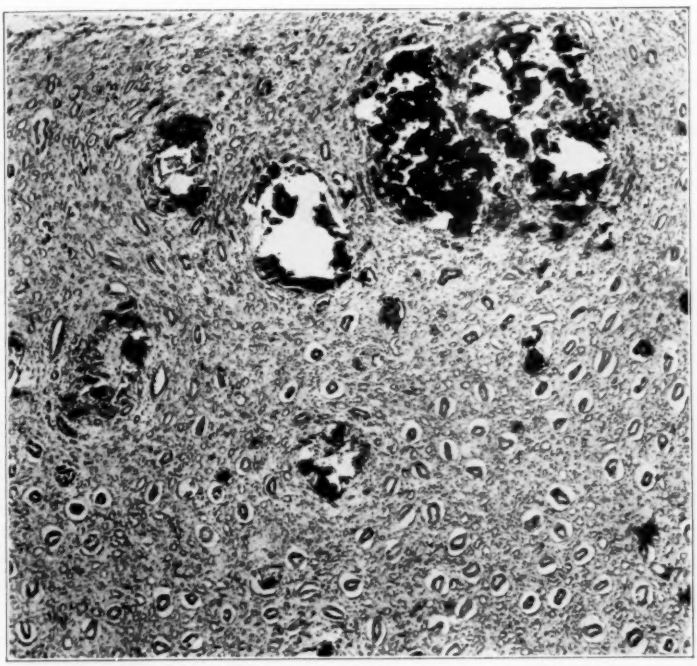


Fig. 7 (case 2).—Kidney with metastatic calcification. Hematoxylin and eosin; × 60.

following values: sugar, 112 mg.; urea, 71.1 mg.; uric acid, 9.6 mg.; total proteins, 4.67 mg.; albumin, 2.26 mg.; globulin, 2.41 mg.; cholesterol, 154 mg.; free cholesterol, 82 mg.; total lipids, 614 mg.; chlorides, 316 mg.; calcium, 15.6 mg.; phosphorus, 3.5 mg.; total base, 140 mg.; sodium, 280 mg., and phosphatase, 8.2 units. The carbon dioxide content was 65.5 volumes per cent. The albumin-globulin ratio was 0.94. The icterus index was 3.6. The Kline reaction was negative. Repeated cultures of the blood gave negative results. The sedimentation rate was 210 mm. per hour.

Examination of bone marrow obtained by aspiration on November 2 showed: myeloblasts, 4; myelocytes, 35; eosinophilic myelocytes, 4; metamyelocytes, 4;

stab forms, 8; polymorphonuclear leukocytes, 30; eosinophils, 5; granulocyte-lobocyte ratio, 65:35; granulocyte-erythroblast ratio, 80:20; ratio of early to later erythroblasts, 25:75, and white blood cells per high power field, 8.

A concomitant smear of the peripheral blood showed: stab forms, 5; polymorphonuclear leukocytes, 59; lymphocytes, 28; monocytes, 4; esosinophils, 4, and platelets, 3 plus. There were anisocytosis (2 plus), microcytosis (3 plus), macrocytosis (2 plus) and polychromasia (2 plus).

By a study of the bone marrow and a smear of the peripheral blood leukemic myelosis was excluded.

Necropsy.—The body was well developed but poorly nourished. Around the outer margins of both breasts the skin was pale brown, leathery and raised above the surface; the condition extended into the axillas and was also present over

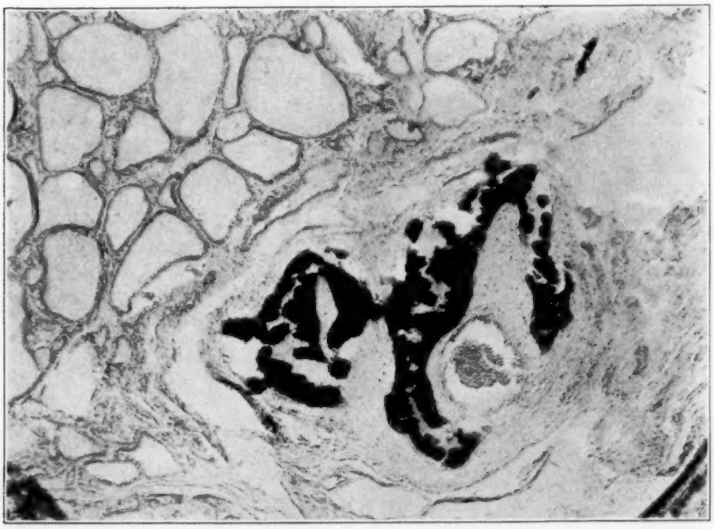


Fig. 8 (case 2).—Thyroid gland with edema and calcium deposits in the wall of the vessel. Hematoxylin and eosin; × 75.

the anterior aspects of both forearms. Within the affected areas were old scars and small raised nodules of cardboard-like consistency. Similar lesions were present over both thighs and legs. There was a large decubitus ulcer over the right greater trochanter. There was edema of the labia majora and of the lower extremities. There was 100 cc. of blood-tinged fluid in the peritoneal cavity, and 1,000 cc. of similar fluid was present in each pleural cavity.

The heart weighed 260 Gm. The right side was not unusual. In the left atrium the endocardium was studded with numerous firm, calcific yellow-white plaques. Similar plaques were also present in the aortic cusp of the mitral valve. The cusps of the aortic valve were somewhat shortened and slightly thickened.

The aortic valve ring was calcified in places. The coronary arteries were widely patent. They cut with a gritty sensation. In the intimal surfaces were

seen numerous small firm yellow plaques. The aorta and pulmonary arteries were elastic and except for occasional atheromatous patches showed nothing of note.

The lungs were heavy, weighing about 600 Gm. each. They did not collapse when the thoracic cavity was opened. The external surfaces were smooth, glistening and mottled with pink and purple-red. The cut surfaces were pink, with occasional raised red areas. There was a granular feel to the cut surfaces. The vessels stood out prominently.

The spleen weighed 190 Gm. It was soft and deep red on section. The malpighian corpuscles and fibrous markings were prominent. There were three small recent infarcts near the surface.

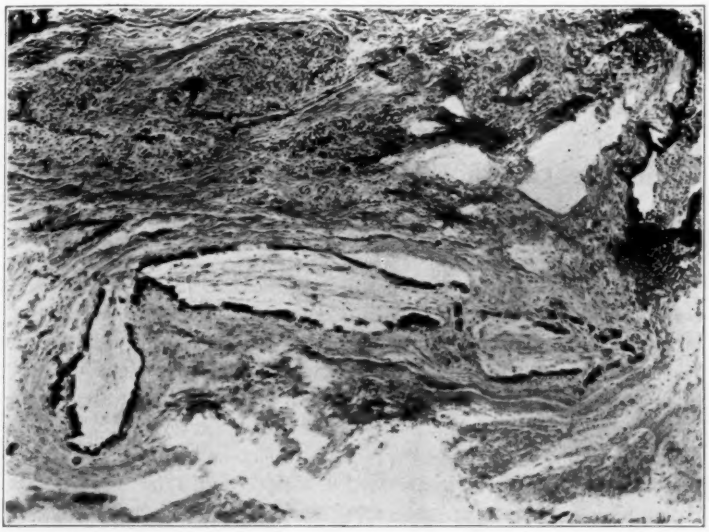


Fig. 9 (case 2).—Mammary gland with vascular changes. Hematoxylin and eosin; × 82.

Grossly there was little of note in the other organs except for marked congestion of the vessels and scattered petechiae. The liver had the typical nutmeg appearance. The kidneys weighed 160 Gm. each. The capsules stripped with ease, leaving smooth surfaces over which were scattered small petechiae. The cortex and medulla were well demarcated. There was evidence of slight cystitis and slight proctitis. Small calcified plaques were present over the vocal cords. The thyroid gland was small and not unusual. The parathyroid glands, of which there were three, were also small.

Microscopic Examination.—The striking picture in the various preparations from the different organs was seen in the veins and smaller arteries. The lumens were somewhat diminished owing to edema of the subintimal fibrous connective tissue. Within the intimae there were fairly extensive deposits of calcium. In some vessels these deposits was also present in the media and even in the

adventitiae. In the lungs, in addition to these vascular changes, there were calcium deposits in the bronchial walls immediately beneath the epithelial layer, in the walls of some alveoli and within the peribronchial and perivascular connective tissue. The lungs also showed areas of pneumonia. In the kidneys calcium deposits were seen not only in the vessels and in the walls of the tubules but also within the tubules and scattered widely through the interstitial connective tissue.

Similar changes were seen in the vessels of the breasts, pancreas, spleen, uterus, cervix uteri and thyroid gland, as well as in the smaller branches of the mesenteric vessels.

In the skin extensive calcium deposits were seen in the midcutis, apparently affecting primarily the elastic tissue fibers.

The parathyroid glands histologically were not unusual.

Analysis of a cutaneous plaque gave the following values: calcium, 114 mg.; phosphorus, 1.6 mg.; total lipid, 0.2813 Gm.; total cholesterol, 0.935 mg., and free cholesterol, 0.62 mg., per gram of tissue. Analysis of normal skin gave the following results: calcium, 0.1 mg.; phosphorus, 1.1 mg., and total lipid, 7.4 mg., per gram of tissue.

All the calculations were done on the dry basis.

The anatomic diagnosis was metastatic calcification in the skin, heart, lungs, spleen, pancreas, kidney, uterus, breast, thyroid and larynx; generalized purpura; focal myocarditis; dilatation of the heart; bilateral hydrothorax; ascites; edema of the labia majora and the lower extremities; passive congestion of the viscera; bilateral focal pneumonia; cystitis; proctitis; infectious splenomegaly; and infarcts of the spleen.

Two general types of pathologic calcification occur in the human body, the dystrophic type, in which the lime salts are deposited in previously diseased tissue, and the other, in which the salts are deposited in apparently healthy tissue. The latter is subdivided by some into calcinosis universalis, in which there is no increase of calcium in the blood, and metastatic calcification, in which there is hypercalcemia. The latter distinction appears to be unnecessary since in both instances the cause of the condition is little understood and since both types are associated with a variety of diseases. Moreover, as in a case cited by Shelling,[4] the concentration of serum calcium in the same patient may vary between normal and tetanic values.

Since no degenerative changes precede the deposition of calcium salts in the various tissues, the cause must be sought in the general factors responsible for calcium and phosphorus metabolism in the body and for demineralization of the skeleton. Some of the suggested causes are disturbances in the vascular supply to the part, disturbances in the calcium and phosphorus metabolism, disturbances in the acid-base equilibrium due to loss of carbon dioxide in the tissues,[5] endocrine

4. Shelling, D. H.: The Parathyroids in Health and in Disease, St. Louis, C. V. Mosby Company, 1935, p. 39.

5. Wells, H. G.: Calcification and Ossification, Arch. Int. Med. **7**:721 (June) 1911; Metastatic Calcification, ibid. **15**:574 (April) 1915.

changes, hypervitaminosis D, infections and trauma. Ham [6] concluded that in metastatic calcificaton there is inability of the blood to retain the calcium in solution. The level of diffusible calcium, therefore, is more important than the level of total calcium.

SUMMARY

Two cases of metastatic calcification are reported. In the first the condition was associated with a leukemic myelosis; in the second it complicated a normal pregnancy in an apparently healthy young woman. The cause was not determined.

6. Ham, A. W.: Mechanism of Calcification in Heart and Aorta in Hypervitaminosis D, Arch. Path. **14**:613 (Nov.) 1932.

Progress in Internal Medicine

BLOOD

A REVIEW OF THE LITERATURE

CYRUS C. STURGIS, M.D.

RAPHAEL ISAACS, M.D.

S. MILTON GOLDHAMER, M.D.

AND

FRANK H. BETHELL, M.D.

ANN ARBOR, MICH.

(Concluded from page 1231)

ANEMIA OF PREGNANCY

Experimental studies of the effects on the blood of iron deficient diets given to rats throughout gestation and lactation have been discussed earlier in this review,[159] A macrocytic form of anemia in pregnant rats has been produced by Kyer and Bethell [160] by means of diets deficient in protein. Such anemia is accompanied by characteristic changes in the percentage and absolute number of the nucleated cells in the several stages of erythrocytic development represented in the marrow. [161] The specific alteration in erythropoiesis is a "shift" to the more immature nucleated red blood cells, analogous to the "megaloblastic block" found in the marrow of patients with pernicious anemia. In rats receiving an inadequate amount of protein during gestation there is also extensive lipoidosis of the liver, arising first in the central zones of the hepatic lobules. An etiologic relation between hepatic damage, induced by deficient diets, and specific changes in the marrow and circulating blood was suggested.

Bussabarger and his associates [162] studied the effects of gastrectomy on the blood values of 5 dogs observed throughout fifteen pregnancies. Marked anemia developed during the course of eleven gestations, and a decrease in total circulating hemoglobin occurred regardless of changes

159. Alt,[137] Parsons, Hickmans and Finch.[141]

160. Kyer, J., and Bethell, F. H.: Production of Macrocytic Anemia in the Pregnant Rat by Diets Low in Protein, Arch. Path. **25**:761 (May) 1938.

161. Bethell, F. H.: Studies of the Bone Marrow and Liver in Experimental Anemia of Pregnancy, Arch. Path. **25**:764 (May) 1938.

162. Bussabarger, R. A.; Cuthbert, F. P., and Ivy, A. C.: Studies on the Anemia of Pregnancy in Gastrectomized and Normal Dogs, J. Lab. & Clin. Med. **24**:24, 1938.

in the total blood volume. The authors also observed the blood values of 12 normal dogs during pregnancy and found severe anemia in only 1 instance. In normal animals, lowering of the erythrocyte and hemoglobin levels was more pronounced in the puerperium than during gestation. Blood volume determinations for 8 dogs disclosed hydremia incidental to pregnancy in only 3 instances. Anemia in pregnancy following gastrectomy was of either normochromic or hypochromic type, and its development was attributed largely to a deficiency of reserve stores of blood-building materials.

Copper and iron determinations for human maternal and fetal blood were reported by Sachs and his co-workers.[163] They found the whole blood iron content of the newborn to be high, whereas the copper value was uniformly low. On the other hand, the maternal blood iron content was normal or reduced, but the copper content was relatively high. The authors pointed out that large amounts of copper are stored in the fetal liver.

The important subject of changes in the blood volume which occur in healthy pregnant women was reinvestigated by Thomson and his associates,[164] who employed in their studies the azo dye Evans blue, and the Evelyn photoelectric microcolorimeter. They found a progressive increase in the plasma and the total blood volume, beginning early in pregnancy and reaching a maximum during the ninth lunar month. During the tenth lunar month a definite decrease in both the plasma and the total blood volume begins, which by the end of the second postpartal week brings the values close to the average normal level for nonpregnant women. The authors found in normal pregnancy a maximum increase in plasma volume of 65 per cent. According to them an absolute increase in the total cell volume also occurs but, proportionately, to a less degree. The increases in the plasma and the total blood volume determined by these workers are considerably greater than those reported by other investigators, and their results await confirmation.

Odefey [165] found a 40 per cent incidence of hemoglobin values below 12 Gm. per hundred cubic centimeters in 103 women observed during pregnancy. Most of the cases he considered as representing "pseudoanemia," since the women were in apparent good health and their blood values, in the absence of therapy, returned to the normal range for non-

163. Sachs, A.; Levine, V. E.; Griffith, W. O., and Hansen, C. H.: Copper and Iron in Human Blood: Comparison of Maternal and Fetal Blood After Normal Delivery and After Cesarean Section, Am. J. Dis. Child. **56**:787 (Oct.) 1938.

164. Thomson, K. J.; Hirsheimer, A.; Gibson, J. G., Jr., and Evans, W. A., Jr.: Studies on the Circulation in Pregnancy: III. Blood Volume Changes in Normal Pregnant Women, Am. J. Obst. & Gynec. **36**:48, 1938.

165. Odefey, R.: Weitere Untersuchungen über die Pseudoanämie in der Schwangerschaft, Zentralbl. f. Gynäk. **62**:1981, 1938.

pregnant women after delivery. Foderl [166] also described "pseudo-anemia" and differentiated it from "essential anemia of pregnancy," which he stated is a true iron deficiency anemia, being responsive to iron therapy. He discussed pernicious anemia of pregnancy, which usually responds well to the administration of liver extract and iron and only rarely requires blood transfusion. A third and rare form of anemia of pregnancy mentioned by this author was described as normocytic and is believed to be of the aplastic type. It is associated with toxemia, and its presence is an indication for blood transfusion. Most observers believe that the rare grave anemias accompanying toxemia are generally hemolytic rather than aplastic. Ramsay and Thierens [167] studied the chemistry of the blood during pregnancy and found hemoglobin values below 9.7 Gm. per hundred centimeters in 26 per cent of their series of about 100 patients. Watson [168] stated that the physiologic lowering of the hemoglobin value in pregnancy may reach 7.7 Gm., but his evidence that these low values are actually normal was inconclusive.

Reid and Mackintosh [169] determined the hemoglobin values for 1,094 women of poor social circumstances and attempted to ascertain the influence of anemia on the subsequent history of the mother and child. Of the women studied, 10.2 per cent suffered from hypochromic anemia, according to the authors' criterion of normality for pregnancy, a minimum hemoglobin level of 9.7 Gm. per hundred centimeters. They found that the incidence of anemia was inversely proportional to the family income. Anemia alone appeared to have a slight influence on the stillbirth rate, and anemia combined with poor social circumstances exerted a definite influence on neonatal and infant death rates and on infant morbidity rates. Mothers who had suffered from anemia during pregnancy were not handicapped in establishing breast feeding but were less able to maintain lactation than nonanemic mothers. No relation was apparent between anemia and toxemia, an abnormality or prolonged duration of labor or the birth weight of the infant.

Napier and Majumdar [170] reported their analyses of the blood and the results of therapy in 57 cases of anemia in pregnant tea-garden coolie

166. Foderl, V.: Zur Differential Diagnose und Therapie der Schwangerschaftsanämien, Wien. klin. Wchnschr. **51**:168, 1938.

167. Ramsay, G., and Thierens, V. T.: The Composition of the Blood in Pregnancy, Brit. M. J. **1**:1199, 1938.

168. Watson, H. G.: The Blood Picture of Pregnancy, Am. J. Obst. & Gynec. **35**:106, 1938.

169. Reid, W. J. S., and Mackintosh, J. M.: The Influence of Anemia and Poor Social Circumstances During Pregnancy on the Subsequent History of Mother and Child, Lancet **2**:1389, 1937.

170. Napier, L. E., and Majumdar, D. N.: Haematological Studies in Indians: The Analysis of the Haematological Findings in Fifty-Seven Cases of Anaemia in Pregnant Tea-Garden Coolie Women, with Special Reference to the Results of Treatment, Indian J. M. Research **26**:541, 1938.

women. The anemia was usually hypochromic or normochromic; 3 patients had a hyperchromic blood picture. Etiologic factors were the universal incidence of hookworm infestation and marked dietary deficiencies in these women. Protein, calcium and vitamins B and C were especially deficient. Treatment consisted of the administration of 1.2 to 1.6 Gm. of iron as ferrous sulfate daily, 4 cc. of autolyzed yeast three times a day and, in the presence of hyperchromic anemia, 2 cc. daily of liver extract by intramuscular injection. Chappell and Bivings [171] studied the blood of 329 pregnant Negresses and compared the findings with those obtained for 75 nonpregnant Negro clinic patients. The hemoglobin range was low, and the color index was reduced in most of the cases studied, but the hemoglobin values for the nonpregnant subjects averaged only 9.0 Gm. per hundred centimeters, and the average level in pregnancy was 8.8 Gm. Poor results were obtained from iron therapy, parenteral administration of liver extract and treatment with supplementary vitamins A and D, and the authors emphasized the inadequacy of the diets of the group investigated.

A series of 100 patients with anemia of pregnancy was reported on by Stevenson.[172] Of these, 30 were said to possess the blood findings of pernicious anemia of pregnancy, 27 of those with pernicious anemia were multiparae and 28 were of a very low economic status. Nutrition was generally inadequate; the symptoms of anemia developed in most instances between the sixth and the eighth month but in 3 cases not until the puerperium. An icteric tinge of the skin was present in 24 of these cases, pyrexia in 19, glossitis in 15, splenomegaly in 15 and achlorhydria in 3. The excretion of urobilinogen was increased in 10 cases. Involvement of the central nervous system was not found. Good therapeutic results, including reticulocyte responses, were obtained from the use of liver, liver extract and desiccated stomach. The 70 other members of the group all suffered from anemia of the hypochromic type and responded satisfactorily to iron therapy.

Cases of severe macrocytic or "pernicious anemia" of pregnancy have also been reported recently by Dockeray,[74] Abramson [173] and Barnum Woodward.[174]

POISONS; PHYSICAL AGENTS

Wood [89] found that of 522 patients treated with sulfanilamide 21 had acute anemia (children, 8.3 per cent; adults, 2.4 per cent). No pre-

171. Chappell, A., and Bivings, L.: Anemia and Pregnancy: A Three Year Study on Negro Women, South. M. J. **31**:876, 1938.

172. Stevenson, E. M. K.: Anaemia in Pregnancy and the Puerperium, Tr. Edinburgh Obst. Soc., 1937-1938, p. 81; in Edinburgh M. J., July 1938.

173. Abramson, L.: Etats rappelant l'anémie pernicieuse durant la grossesse et les suites de couches, Acta med. Scandinav. **96**:319, 1938.

174. Barnum, C. G., and Woodward, J. C.: Severe Hyperchromic Macrocytic Anemia of Pregnancy, J. A. M. A. **111**:1749 (Nov. 5) 1938.

disposing factors could be determined. The earliest signs of anemia were noted from twenty-three to seventy-two hours after the start of the therapy. The anemia was worst usually on the fifth day (third to seventh day). It was hemolytic and recurred in 4 of 5 patients during a second course of sulfanilamide therapy.

Campbell [175] did not encounter anemia or depression of the bone marrow in his series of patients treated with sulfanilamide, and concluded that "personal idiosyncrasy" is an important factor in the cases of involvement of the bone marrow reported by others.

The case reported by Ginsberg and Brams [176] illustrated the rapidity with which hemolytic anemia secondary to sulfanilamide therapy can develop. After four days of therapy (160 grains [10.4 Gm.]), icterus was noted, and the red blood cell count on the seventh day had fallen from 4,500,000 to 1,100,000 red blood cells per cubic millimeter (hemoglobin, 25 per cent; leukocytes, 36,900 per cubic millimeter; neutrophils, 66 per cent). Recovery followed three blood transfusions (500 cc. each) on successive days. Nelson and Scott-Young [177] and Wood [89] also reported cases in which hemolytic anemia followed sulfanilamide therapy.

In some persons who were cyanotic from sulfanilamide therapy, Posner, Guthrie and Mattice [178] found an abnormal blood pigment, having the same absorption band as methemoglobin. The band disappeared on saturation with carbon monoxide or dilution with water. The pigment was unstable, slowly reverting to normal blood pigment.

In patients treated for fourteen days with 21 Gm. of sulfanilamide, Britton and Howkins [179] found that 46 per cent showed neutropenia and 44 per cent transient monocytosis. The lowest count was noted at the end of the second or at the beginning of the third week. However, the toxic symptoms, present in 70 per cent of the patients, showed no direct relation to the variation in the leukocyte count.

Weil, Perlés and Aschkenasy [180] studied the blood of 54 patients with anemia of benzene poisoning. The blood characteristics were hypochromia, leukopenia, neutropenia, eosinophilia (4 to 8 per cent in 30

175. Campbell, C. M.: Effect of Sulfanilamide on the Blood Picture, Lancet **1**:247, 1938.

176. Ginsberg, A. M., and Brams, J. B.: Acute Hemolytic Anemia Following Treatment with Sulfanilamide, J. Missouri M. A. **35**:174, 1938.

177. Nelson, E. L., and Scott-Young, M.: Profound Anemia Following Administration of Sulfanilamide in Acute Gonorrhea, M. J. Australia **1**:626, 1938.

178. Posner, I.; Guthrie, N. W., and Mattice, M. R.: Formation of Abnormal Blood Pigment as Complication of Sulfanilamide Therapy, J. Lab. & Clin. Med. **23**:804, 1938.

179. Britton, C. J. C., and Howkins, J.: Action of Sulfanilamide on Leukocytes: Report of Fifty Ambulant Patients, Lancet **2**:718, 1938.

180. Weil, P. E.; Perlès, S., and Aschkenasy, A.: Etudes sur le benzolisme latent, Sang **12**:151, 1938.

per cent of the cases), thrombopenia, clotting defects and capillary fragility. The latent form is an aplastic anemia. The authors made a plea for well ventilated rooms, limited hours of work and regular medical inspection of workers in benzene.

In a case of benzene poisoning, Gall [181] found aplastic anemia, leukopenia, thrombopenia and macrocytosis. The peripheral circulation, however, showed reticulocytes, nucleated red blood cells and immature myeloid cells, secondary to massive myeloid (erythropoietic) metaplasia in the spleen and, to a less extent, in the liver. Zolezzi [182] found decreased gastric function accompanying anemia in 8 cases of benzene poisoning in workers in the rubber industry. He also noted several cases of anemia among leather workers in shoe factories due to adhesives and mastics dissolved in benzene. In Kern's [183] case of benzene poisoning, there was a selective reduction in the number of blood platelets, with disappearance from the blood. Later there was an increase to above normal, but the red and white blood cells did not appear to be involved in the process. Neurologic symptoms, presumably secondary to hemorrhage into the central nervous system, were present. Tara [184] described chronic benzene poisoning in 14 pregnant women. Of these, 4 had normal deliveries; 3 had difficult deliveries, with uterine inertia; 1 gave birth to a dead child, and 7 had abortions.

Rajam and Tampi [185] noted 3 cases of blood dyscrasia after treatment with arsphenamine and similar agents in 18,620 cases of syphilis (64,-101 injections). One patient had thrombopenic purpura (platelets, 50,000). The second patient had aplastic anemia and the third granulocytopenia. Treatment included the use of epinephrine, calcium, thiosulfate, liver preparations, pentnucleotide and repeated transfusions. Arsenical injections must be discontinued, as recurrences are invariably fatal. Kadin [186] reported 3 cases of aplastic anemia with severe damage to the bone marrow in patients who had been treated with arsphenamine. All the patients showed a hemorrhagic tendency.

In pregnant female rats Briese [187] produced anemia by means of inhalation of carbon tetrachloride. Macrocytic anemia, with an increased

181. Gall, E. A.: Benzene Poisoning with Bizarre Extramedullary Hematopoiesis, Arch. Path. **25**:315 (March) 1938.

182. Zolezzi, G. G.: Alterazione del chimismo gastrico nella intossicazione da benzolo, Med. d. lavoro **28**:275, 1937.

183. Kern, B.: Isolierte-Thrombopenie durch chronische Benzolvergiftung, München. med. Wchnschr. **85**:1062, 1938.

184. Tara, S.: Le benzénisme larvé chez la femme enceinte, Sang **12**:352, 1938.

185. Rajam, R. V., and Tampi, R. B.: Postarsphenamine Blood Dyscrasias, Indian M. Gaz. **73**:337, 1938.

186. Kadin, M.: Aplastic Anemia Following the Use of Neoarsphenamine, Arch. Dermat. & Syph. **37**:787 (May) 1938.

187. Briese, E.: The Effect on the Blood Cells of the Fetal Rat Produced by the Inhalation of Carbon Tetrachloride by the Mother During Gestation, Am. J. M. Sc. **195**:787, 1938.

hemoglobin content, reticulocytosis, thrombocytosis, leukocytosis and an increased immaturity of the leukocytes, developed in the fetus.

In the course of gold therapy in pulmonary tuberculosis, Daniello and Rusu [188] noted an increase in the hemoglobin content and in the eosinophil and lymphocyte counts. Some patients showed purpura, agranulocytosis and anemia.

Shiraishi [189] found that yakriton (a fraction of liver not containing the principle potent against pernicious anemia) alleviated phenylhydrazine poisoning or the severe anemia which it causes in rabbits.

Loureau, de Sacy and Arthus [190] were able to produce a prolonged period of anemia in rabbits by the injection of lead salts three times a week for three weeks.

Ammonia, administered to rabbits, produced a reduction in the number of red blood cells and the hemoglobin content of the blood in the experiments of von Baló.[191] The anemia was proportional to the degree of poisoning. The cellular elements of the red bone marrow were destroyed first and later the myeloid (polynuclear) elements, with an increase in the number of lymphoid cells.

Kornblum, Boerner and Henderson [192] studied before and after roentgen therapy certain features of the blood of 48 patients who did not have outstanding disease of the hemopoietic organs. After treatment, over half the patients showed a decrease in the red blood cell count and volume index, over 83 per cent showed a decrease in the leukocyte count, over 72 per cent showed a decrease in the number of neutrophils and in the platelet count, and over 81 per cent showed a decrease in the number of lymphocytes. The authors concluded that roentgen irradiation "depresses" the bone marrow and slows the production of cells. This, however, does not explain the fact that after irradiation many patients in their series showed just the opposite effect, namely, an increase in numbers. Yet they feel "no stimulating effects were observed." The authors [193] studied the blood of 100 patients with various types of cancer and lymphoblastoma treated with a wide range of doses of roentgen rays

188. Daniello, L., and Rusu, V.: Hämatologische Veränderungen im Laufe der Goldtherapie, Beitr. z. Klin. d. Tuberk. **89**:352, 1937.

189. Shiraishi, S.: Studies on the Detoxicating Hormone of the Liver (Yakriton): Effect of Yakriton upon Phenylhydrazin Anemia in Animals of Low-Classed Liver Power, Tohoku J. Exper. Med. **32**:338, 1938.

190. Loureau, M.; de Sacy, G. S., and Arthus, A.: Stabilité de l'anémie produite dans le saturnisme expérimental, Compt. rend. Soc. de biol. **128**:512, 1938.

191. von Baló, J.: Der Einfluss der Ammoniakvergiftung auf das Blut und auf die Blutbildung, Beitr. z. path. Anat. u. z. allg. Path. **101**:66, 1938.

192. Kornblum, K.; Boerner, F., and Henderson, S. G.: The Qualitative Changes in the Formed Elements of the Blood Following Therapeutic Irradiation. Am. J. Roentgenol. **39**:601, 1938.

193. Kornblum, H.; Boerner, F., and Henderson, S. G.: The Effects of Irradiation on the Normal Blood Cells as Determined by the Blood Count, Am. J. Roentgenol. **39**:235, 1938.

or radium. They concluded that with therapeutic doses there was no significant effect on the red blood cell count or the hemoglobin percentage and that anemia per se was not a contraindication to radium therapy. The greatest decrease occurs in the lymphocyte and then in the neutrophil count, while the monocyte and eosinophil counts are least affected. The part of the body treated and the dosage appeared to be minor factors compared with the size of the area treated. The leukocyte count usually returns to normal in five weeks; if it is prolonged to eight weeks the prognosis is unfavorable. Liver extract did not prevent the reduction in the leukocyte count or restore it to normal after treatment. Irradiation did not predispose to infection. "From a practical point of view," the authors concluded, "the effects of irradiation on normal blood, as determined by the blood count, are of little clinical significance." Furth, Tuggle and Breedis,[194] in experimenting with the action of x-rays on neoplastic cells, found that atypical cells of monoculear leukemia survived exposure to 5,000 roentgens but could not reproduce the disease after exposure to 15,000 roentgens. Virus diseases survive this dosage of roentgen rays, suggesting that neoplasms are not virus diseases. After irradiation, the cells do not die at once but reproduce for several generations (in vitro) before they degenerate and die. Yamashita[195] found a marked reduction in the number of leukocytes and lymphocytes in rats irradiated with neutrons, although the neutrophils were increased in number. Eosinophils disappeared quickly; the monocytes were first slightly increased in number and later slightly decreased. There was a slow decrease in the number of erythrocytes and in the hemoglobin content. Anemia was evident in all the organs, with a reduction in the size of the spleen, testis, ovary and bones.

In tumor-bearing animals, Clarkson, Mayneord and Parsons[196] noted profound anemia after irradiation. Iron deposits were found in the spleen, fatty changes were present in the liver and kidneys and the amount of lymphoid tissue was reduced.

Doan[197] found that during treatment with artificial fever, there was polymorphonuclear neutrophilia (newly delivered cells). There were destruction of lymphocytes and delayed monocytosis (younger forms). In induced pyrexia with malaria treatment or Bacillus typhosus, there is marked leukopenia during the chill. In typhoid pyrexia there is a

194. Furth, J.; Tuggle, A., and Breedis, C.: Quantitative Studies on the Effect of X-Rays on Neoplastic Cells, Proc. Soc. Exper. Biol. & Med. **38**:490, 1938.

195. Yamashita, H.: Experimentelle Untersuchungen über die biologischen Wirkungen der Neutronen, Gann **31**:654, 1937; abstracted, Biological Effects of Deuteron-Deuteron-Neutrons, Nature, London **141**:416, 1938.

196. Clarkson, J. R.; Mayneord, W. V., and Parsons, L. D.: The Effect of X-Radiation on the Blood and Lymphoid Tissue of Tumor Bearing Animals, J. Path. & Bact. **46**:221, 1938.

197. Doan, C. A.: Peripheral Blood Phenomena and Differential Response of Bone Marrow and Lymph Nodes to Hyperpyrexia, Radiology **30**:382, 1938.

temporary disappearance of the peripheral monocytes, but a moderate stimulation with typhoid vaccine and a marked stimulation from malaria treatment occur. In the latter type, clasmatocytes appear in the peripheral blood.

Bensley, Rhea and Mills [198] described the cases of a brother and sister with familial idiopathic methemoglobinemia. Only 5 cases had been recorded previously. Methemoglobin was identified spectroscopically, and the content of oxygen-carrying hemoglobin was less than the total hemoglobin content. No extraneous cause for the cyanosis was found.

Minot [199] has reviewed the effects of lead on the growth of plant and animal tissue, enzyme activity, formation of immune bodies, red blood cells, white blood cells, skeletal and smooth muscle and nervous and other systems.

THE BLOOD IN DISEASE OF THE KIDNEYS

Löwinger [200] studied azotemic anemia (nephritis) by means of bone marrow puncture. The anemia is of the nonregenerative type, with inhibition of the maturation of the erythroblasts by a toxic substance.

Ishida [201] found that anemia followed glomerular injury with reagents but not when the tubules were damaged. The anemia-producing factor was insoluble in water, but it dissolved in lipoid solvents and was saponifiable

Drury [202] produced renal insufficiency in rabbits by limiting the blood flow in one kidney and later removing the other. Moderate anemia developed rapidly after the reduction of the total renal mass. Hemorrhage into the wall and lumen of the intestine was noted in many of the animals.

From roentgenograms of the bone marrow of patients with chronic nephritis, Nordenson [203] concluded that there was a mild degree of aplasia of the precursors of red blood cells, without involvment of the white blood cells. The number of nucleated red blood cells was decreased.

---

198. Bensley, E. H.; Rhea, L. J., and Mills, E. S.: Familial Idiopathic Methaemoglobinaemia, Quart. J. Med. **7**:325, 1938.

199. Minot, A. S.: The Physiological Effects of Small Amounts of Lead: An Evaluation of the Lead Hazard of the Average Individual, Physiol. Rev. **18**:554, 1938.

200. Löwinger, S.: Die Erythropoese bei azotämischen Anämien, Folia haemat. **59**:222, 1938.

201. Ishida, K.: A Supplement to "The Anemic Factor in Nephritis," J. Chosen M. A. **27**:1506, 1937.

202. Drury, D. R.: The Production by a New Method of Renal Insufficiency and Hypertension in the Rabbit, J. Exper. Med. **68**:693, 1938.

203. Nordenson, N. G.: Bone Marrow in Anemia of Chronic Nephritis, Folia haemat. **59**:1, 1938.

POLYCYTHEMIA

Haden [204] pointed out that whereas the total blood volume is constantly increased in polycythaemia vera, the plasma volume is seldom greater than normal. The red bood cell mass per kilogram is the most sensitive indicator of the changes in the red blood cells. The red blood cell mass is constantly high in polycythaemia vera but not significantly changed in symptomatic polycythemia. The total mass of red blood cells is a guide for the control of treatment.

Hallock [205] found that whereas in normal persons the average lactic acid concentration of the venous blood during rest was 13.9 mg. per hundred centimeters and 16.3 mg. after mild exercise, in polycythaemia vera the values were 17.8 and 15.1 mg., respectively. The same response to exercise, however, was found after reducing the red blood cell count and hemoglobin percentage to normal, suggesting that the disturbance of lactic acid metabolism in polycythaemia vera was dependent on some primary factor, to which the polycythemia itself was secondary. In the polycythemia of congenital heart disease the figures were 17.5 and 32.2 mg. per hundred centimeters, respectively, and in marked emphysema the numbers were 19.8 and 22.2 mg.

Glasser [206] described the successful treatment of a 60 year old polycythemic woman with phenylhydrazine hydrochloride. Splenomegaly and hepatomegaly were unusually marked. Three of the patient's brothers and nephews had hemophilia.

Di Guglielmo [207] described acute erythremic myelosis (acute erythremia) as a clinical entity. It is characterized by severe anemia, irregular or remittent fever, splenomegaly, moderate hepatomegaly, the presence of basophilic erythroblasts in the blood, primary hyperplasia of the erythropoietic tissue, proliferation of the cells of the reticuloendothelial system, a duration of from one to two months and a fatal outcome. Incomplete and atypical forms are encountered. The disease occurs in both sexes and at all ages. Therapeutic efforts (liver, arsenic, an iron preparation or blood transfusions) are unavailing.

Rodes [208] found polycythemia (red blood cells, 7,540,000; hemoglobin, 118 per cent) in a 25 year old man with multiple hemangioma of the lung.

204. Haden, R. L.: The Red Cell Mass in Polycythemia in Relation to Diagnosis and Treatment, Am. J. M. Sc. **196**:493, 1938.

205. Hallock, P.: Blood Lactic Acid After Exercise, with Particular Reference to Polycythemia Rubra Vera, Proc. Soc. Exper. Biol. & Med. **38**:587, 1938.

206. Glasser, S. M.: A Case of Polycythemia Vera: The Familial Incidence of Hemophilia and Treatment with Phenylhydrazine, J. A. M. A. **110**:2071 (June 18) 1938.

207. di Guglielmo, G.: L'érythrémie aiguë, Rev. belge sc. méd. **10**:200, 1938.

208. Rodes, C. B.: Cavernous Hemangiomas of the Lung with Secondary Polycythemia, J. A. M. A. **110**:1914 (June 4) 1938.

Clinically there were cyanosis of the lips, clubbing of the fingers and toes, jaundice, hemangiomas (small) of the lips and a history of hemoptysis. Death resulted from rupture of a hemangioma into the bronchus.

Stone and Woodman [209] described a patient with polycythaemia vera in whom "leukoerythroblastic" anemia developed. The value for nucleated red blood cells in the peripheral blood rose to 84,000 and that of the white blood cells to 94,500 per cubic millimeter. Death resulted from cardiac failure. There was megakaryocytic hyperplasia of the upper two thirds of the femoral marrow with myelosclerosis below. Multiple foci of extramedullary hemopoiesis were found. Lymph glands showing caseous foci with acid-fast bacilli were present.

Cases illustrating the association of polycythemia and peptic ulcer were described by Sansone [210] and Schneiderbaur.[211]

Davis [212] found that liver feeding reduced the polycythemia caused in dogs by exercise or cobalt chloride, in spite of the continuation of these agents. On cessation of the liver therapy, the polycythemia returned. Stomach U. S. P. and liver extract by mouth were not effective, although one kind of liver extract given by daily intramuscular injections produced the same effect as whole liver in reducing the blood count. Major,[213] however, found that liver extract produced no specific effect when given to patients with polycythaemia vera.

Evesen [214] considers that the action of phenylhydrazine in polycythaemia vera is peripheral, the bone marrow being stimulated, not depressed. The same dosage may vary as to its effects on different patients. The first course of treatment should be given with the patient under hospital observation. In patients over 60 years of age the reaction is more rapid and severe, especially if arteriosclerotic or visceral disease is present.

McAlpin and Smith [215] analyzed the cases of 14 patients with polycythaemia vera treated with acetylphenylhydrazine. Of these, 9 were helped; 2 others, with peptic ulcer, were helped only temporarily. Three patients died. Two necropsies did not suggest toxicity of the drug; the

209. Stone, D. M., and Woodman, D.: Polycythaemia Terminating in Leuco-Erythroblastic Anaemia, J. Path. & Bact. **47**:327, 1938.

210. Sansone, L.: Osservazioni e considerazioni sulle poliglobulie, Arch. per le sc. med. **64**:451, 1937.

211. Schneiderbaur, A.: Polycythaemia vera und Ulcus duodeni, Ztschr. f. klin. Med. **133**:474, 1938.

212. Davis, J. E.: The Reduction of Experimental Polycythemias by Liver Administration, Am. J. Physiol. **122**:397, 1938.

213. Major, R. H.: The Effect of Liver Extract on Polycythemia Vera, J. Lab. & Clin. Med. **24**:65, 1938.

214. Evesen, O. K.: The Action of Phenylhydrazine in Polycythaemia Vera, Acta med. Scandinav., 1938, supp. 90, p. 288.

215. McAlpin, K. R., and Smith, K. E.: Polycythemia Vera: Report of Fourteen Cases Treated with Acetylphenylhydrazine, New York State J. Med. **38**:101, 1938.

third patient probably had Ayerza's disease. One patient became jaundiced, and 2 showed a susceptibility to the drug in usual doses. The authors concluded that acetylphenylhydrazine is probably more effective and less toxic than phenylhydrazine hydrochloride.

Kandel and LeRoy [216] found that ascorbic acid had no influence on the red blood cell counts and hemoglobin levels of patients with polycythaemia vera.

Sohval [217] described a case of polycythemia in which hepatic enlargement suddenly appeared and ascites and jaundice developed rapidly, owing to thrombosis of the hepatic veins. Hepatic enlargement was found to be moderate or marked in 30 of 60 cases of polycythemia. Marked enlargement of the spleen often indicated a complication, such as phenylhydrazine jaundice, leukemic transformation, myocardial decompensation, hepatic cirrhosis or thrombosis of the hepatic veins.

Rosenthal and Bassen [218] reviewed 13 cases of polycythemia, illustrating various courses and terminations. The blood picture may become leukemic, thrombocythemic or anemic or may show combinations of these phases.

PURPURA HAEMORRHAGICA

Bleeding may result from a decrease in the number of platelets, alterations in the plasma or changes in the permeability of the capillaries. The etiologic factors which produce these changes are manifold and have resulted in the description of various types of purpura. A clinical classification according to the complexities of the physiopathologic process and the multiple etiologic factors is of somewhat doubtful value.

Torrioli and Puddu [219] made extracts from spleens of patients with thrombopenic purpura. When this material was mixed with cultures of guinea pig bone marrow, marked injury to the megakaryocytes occurred. The authors were unable to identify this thrombopenic substance. They stated that it was also present in extracts made from a normal spleen, liver, lung, red muscle and lymphatic material but in lesser amounts. They were able to detect its presence in the blood of the splenic vein but not in that of the artery. Similar observations were made by Troland

216. Kandel, E. V., and LeRoy, G. V.: Note on the Lack of Hemoregulatory Effect of Ascorbic Acid on Patients with Polycythemia Vera, Am. J. M. Sc. **196**:392, 1938.

217. Sohval, A. R.: Hepatic Complications in Polycythaemia Vera, with Particular Reference to Thrombosis of the Hepatic and Portal Veins and Hepatic Cirrhosis, Arch. Int. Med. **62**:925 (Dec.) 1938.

218. Rosenthal, N., and Bassen, F. A.: Course of Polycythemia, Arch. Int. Med. **62**:903 (Dec.) 1938.

219. Torrioli, M., and Puddu, V.: Recent Studies on the Pathogenesis of Werlhof's Disease, J. A. M. A. **111**:1455 (Oct. 15) 1938.

and Lee,[220] with the exception that they could not identify or isolate this substance in extracts from normal or other types of diseased spleens.

Purpura may be congenital or acquired. Denninger [221] reported 2 cases of the fulminating type occurring in the same family. From the information given, his conclusions are questionable. Lymphoid hyperplasia associated with thrombopenic purpura was described by Cooley.[222] Toxic purpura complicating scarlet fever was observed by Fox and Enzer.[223] A similar type of purpura was noted by Stoesser and Lockwood [224] in varicella, and by Welt and Kasnetz [225] in rubella. Purpura resulting from drug poisoning may be due to a decrease in the platelet count or capillary injury. Hill,[226] Joekes,[227] Moody [228] and Hoffman, Kahn and Fitzgibbon [229] all reported cases of purpura following the use of sedormid (allylisopropylacetylcarbamide). Allergic purpura due to sensitivity to the toxin of hemolytic streptococci was observed by Ellis.[230] Dutton [231] noted allergic purpura following the ingestion of citrus fruits. Purpura due to benzene poisoning was noted by Kern.[183]

The symptoms associated with purpura are varied, depending on the location and severity of the hemorrhages, the cause of the condition, and

---

220. Troland, C. E., and Lee, F. C.: A Preliminary Report on a Platelet-Reducing Substance in the Spleen of Thrombocytopenic Purpura, Bull. Johns Hopkins Hosp. **62**:85, 1938; Thrombocytopen: A Substance in the Extract from the Spleen of Patients with Idiopathic Thrombocytopenic Purpura that Reduces the Number of Blood Platelets, J. A. M. A. **111**:221 (July 16) 1938.

221. Denninger, H. S.: Purpura Hemorrhagica Fulminans, Southwest. Med. **22**:8, 1938.

222. Cooley, L. E.: Lymphoid Hyperplasia Associated with Thrombopenic Purpura, Arch. Path. **26**:390 (July) 1938.

223. Fox, M. J., and Enzer, N.: A Consideration of the Phenomenon of Purpura Following Scarlet Fever, Am. J. M. Sc. **196**:321, 1938.

224. Stoesser, A. V., and Lockwood, W. W.: Varicella Complicated with Acute Thrombocytopenic Purpura and Gangrene, J. Pediat. **12**:641, 1938.

225. Welt, B., and Kasnetz, J.: Thrombopenic Purpura as a Complication: Association with Acute Mastoiditis, Lateral Sinus Thrombosis, Streptococcus Viridans Bacteremia and Multiple Metastatic Abscesses, Arch. Otolaryng. **27**: 732 (June) 1938.

226. Hill, D. B.: Thrombopenic Purpura Following Allyl-Isopropyl-Acetyl-Carbamide (Sedormid), J. A. M. A. **111**:1459 (Oct. 15) 1938.

227. Joekes, T.: Purpura Haemorrhagica (Werlhof) After Taking Sedormid, Lancet **2**:305, 1938.

228. Moody, A. M.: Thrombocytopenic Purpura Following Use of Allyl-Isopropyl-Acetyl-Carbamide (Sedormid), J. A. M. A. **110**:726 (March 5) 1938.

229. Hoffman, A. M.; Kahn, J., and Fitzgibbon, J. P.: Thrombocytopenic Purpura Following Allyl-Isopropyl-Acetyl-Carbamide (Sedormid), J. A. M. A. **110**:725 (March 5) 1938.

230. Ellis, R. W. B.: Allergic Purpura, Proc. Roy. Soc. Med. **31**:768, 1938.

231. Dutton, L. O.: Thrombopenic Purpura Due to Food Allergy: Case Report, J. A. M. A. **111**:1920 (Nov. 19) 1938.

any existing disease which may be present. Cook [232] commented on the complications arising from bleeding in persons who require dental care. The erythema crises of the visceral type of involvement have been emphasized by Poll.[233] This author pointed out that hemorrhage is more common in males, especially in adolescent and early adult life. An interesting observation was made by Abbott,[234] who studied the course of infection in a case of purpura before and after splenectomy. Preceding the operation, the platelet count was decreased. After operation, in the presence of infection, the platelet count increased, as did the white blood cell count, and decreased as the infection subsided.

As the cause of purpura cannot be determined in every instance, treatment frequently results in failure. Further, since remissions are not uncommon, an evaluation of any therapeutic procedure is difficult. Doan [235] recommended splenectomy in purpura with a platelet deficiency. Aitken [236] observed a patient with thrombopenic purpura whose platelet count remained at a low level for at least one year after operation. Fleischhacker [237] suggested the early use of kephrine hydrochloride (methylaminoacetopyrocatechol hydrochloride) but recommended splenectomy for chronic purpura. Roentgen therapy has been employed by innumerable investigators. Jones, Tocantins and Smith [238] believe it beneficial in certain selected cases of purpura. Garland [239] suggested its use in various other hemorrhagic disorders, such as uterine bleeding, epistaxis, gastric oozing, chlorosis and melena neonatorum. Autohemotherapy has been successfully used by Saxon.[240] Imerman and Imerman [241] were able to control the hemorrhages in purpura by means of large doses of snake venom. The therapeutic effect of vitamin P has

232. Cook, T. J.: Blood Dyscrasias from a Dental Point of View, Am. J. Orthodontics **24**:687, 1938.

233. Poll, D.: Erythema Crises of the Visceral Group, J. Mt. Sinai Hosp. **4**: 997, 1938.

234. Abbott, K. H.: Thrombocytopenic Purpura Hemorrhagica, California & West. Med. **48**:332, 1938.

235. Doan, C. A.: Pathologic Physiology of the Spleen: Rationale of Splenectomy in Congenital Hemolytic Icterus, Thrombocytopenic Purpura and Early Banti's Disease, Northwest Med. **37**:61, 1938.

236. Aitken, G. T.: Review of a Case of Thrombocytopenic Purpura Treated by Splenectomy, J. Michigan M. Soc. **37**:628, 1938.

237. Fleischhacker, H.: Zur Therapie der thrombopenischen Purpura mit Stryphnon, Wien. klin. Wchnschr. **51**:449, 1938.

238. Jones, H. W.; Tocantins, L. M., and Smith, R. M.: Splenic Irradiation in the Treatment of Purpura Hemorrhagica, Ann. Int. Med. **11**:1311, 1938.

239. Garland, L. H.: Roentgen Treatment of Certain Hemorrhagic Disorders, California & West. Med. **49**:123, 1938.

240. Saxon, L.: Studies in Autohemotherapy: Report of a Case of Purpura Rheumatica with New Method of Treatment, Illinois M. J. **74**:191, 1938.

241. Imerman, C. P., and Imerman, S. W.: Thrombocytopenic Purpura Hemorrhagica: Treatment with Massive Doses of Moccasin Snake Venom, California & West. Med. **48**:335, 1938.

been carefully studied by Jersild,[242] It is his opinion that the vitamin is specific in the Schönlein-Henoch purpuras.

Since spontaneous cures are not uncommon, a conservative attitude must be adopted concerning the various therapeutic agents recommended. Elliott [243] pointed out that the suction test for capillary resistance in purpura is most helpful. Although it is not infallible, it appears to be a better index of bleeding than the platelet count and is affected earlier after splenectomy, thus serving as a valuable aid in determining the effect of treatment.

HEMOPHILIA

Although hemophilia was recognized in early biblical times, Otto, in 1803, was the first accurately to describe this disease. The essential characteristics are its inheritance, the occurrence in males, transmission by females, a history of repeated hemorrhages, a prolonged clotting time and a normal bleeding time. Riddell and Haldane [244] have emphasized the importance of the hereditary aspect of the disease. The familial incidence of hemophilia has also been stressed by Glasser.[206]

Because of the disturbance in the clotting mechanism of the blood, complications due to hemorrhages are extremely common. It was previously pointed out that about 78 per cent of hemophiliacs have orthopedic complications and approximately 60 per cent have permanent deformities. A case of hemophilia with a fatal spontaneous hemorrhage from a pharyngeal abcess has been reported by Laurent.[245] The seriousness of abnormal bleeding as a surgical risk has been reviewed by Mertz and Meiks.[246] These authors cited a case of fatal nephrectomy in which the bleeding could not be controlled by transfusions or any other known methods. It is the impression of these investigators that the clotting time cannot be a satisfactory criterion of the clinical course.

There is no known specific therapy. Clark and his co-workers [247] described a preparation made from egg white which has proved satisfactory in reducing the clotting time when given parenterally. Pohle

242. Jersild, T.: Therapeutic Effect of Vitamin P in Schönlein-Henoch Purpura, Lancet **1**:1445, 1938.

243. Elliott, R. H. E.: The Suction Test for Capillary Resistance in Thrombocytopenic Purpura, J. A. M. A. **110**:1177 (April 9) 1938.

244. Riddell, W. J. B., and Haldane, J. B. S.: A Haemophilic and Colour-Blind Pedigree, J. Genetics **36**:45, 1938.

245. Laurent, L. J. M.: Haemophilia with Fatal Spontaneous Haemorrhage from Pharyngeal Abscess, Brit. J. Child. Dis. **34**:282, 1937.

246. Mertz, H. O., and Meiks, L. T.: The Hemophiliac as a Surgical Risk: Report of a Case of Nephrectomy with Death, Urol. & Cutan. Rev. **42**:557, 1938.

247. Clark, G. A.; Gaunt, R. T., and Timperley, W. A.: Some Observations on Blood Clotting, Quart. J. Exper. Physiol. **28**:149, 1938.

and Taylor [248] have demonstrated that a globulin substance derived from beef plasma is most effective in controlling hemorrhages in hemophiliacs when applied locally. However, it is of little value when taken orally. These same authors [249] have also shown that normal or lyophilized plasma contains some unknown substance which aids in the reduction of the clotting time. The factor is not present in globulin. Lawson and Graybeal [250] recommended frequent venesection to reduce the bleeding tendency in hemophiliacs, as it is their opinion that increased venous pressure and damaged capillaries may be contributing factors to the hemorrhage.

BLOOD CLOTTING

The various theories concerned with clot formation have been reviewed by Magath,[251] who also stressed the importance of the role of vitamin K in the development of a blood clot and its relation to prothrombin. The historical development, research problems and chemistry of vitamins K and P have been summarized by McCay.[252] The best sources of vitamin K, as pointed out by Osterberg,[253] are hog liver oil, cabbage, spinach, tomatoes and alfalfa. Dam and Glavind [254] have investigated the vitamin K content of various plants by the curative technic. The richest sources are green leaves, chestnut leaves being the most potent. The strength of the vitamin was only slightly altered when the leaves were withering, and the vitamin was equally distributed throughout the leaves, especially in those with alternating yellow and green areas. Flowers, roots and seeds were poor sources. The lower plants had considerably less vitamin K than the higher plants. Germination in the light enabled peas to synthesize a considerable amount of the vitamin, but the same plant synthesized only a limited amount in the dark.

248. Pohle, F. J., and Taylor, F. H. L.: The Use of a Globulin Substance Derived from Beef Plasma as a Local Hemostatic in Hemophilia, J. Clin. Investigation **17**:677, 1938.

249. Pohle, F. J., and Taylor, F. H. L.: The Coagulation Defect in Hemophilia: Studies on the Refractory Phase Following Repeated Injections of Globulin Substance Derived from Normal Human Plasma in Hemophilia, J. Clin. Investigation **17**:779, 1938.

250. Lawson, G. B., and Graybeal, A. B.: Hemophilia Treated by Venesection, J. A. M. A. **111**:2104 (Dec. 3) 1938.

251. Magath, T. B.: Coagulation of Blood with Special Reference to Prothrombin, Proc. Staff Meet., Mayo Clin. **13**:67, 1938.

252. McCay, C. M.: Other Factors: Less Well Known Vitamins, J. A. M. A. **110**:1441 (April 30) 1938.

253. Osterberg, A. E.: Vitamin K—Its Distribution and Chemical Properties: Methods of Preparation and Assay, Proc. Staff Meet., Mayo Clin. **13**:72, 1938.

254. Dam, H., and Glavind, J.: Vitamin K in the Plant, Biochem. J. **32**:485, 1938.

Almquist and his co-workers [255] were able to demonstrate the presence of vitamin K by assay methods in materials acted on by various bacteria. They suggested that the vitamin was a product of bacterial metabolism.

As pointed out by Osterberg, vitamin K is fat soluble, and nonnitrogenous, has an aromatic nucleus, but contains no phosphorus or sulfur. It has no sterol ring and is alkali labile, heat stable and optically inactive, with a molecular weight of 600.

An accurate method of estimating the prothrombin level and of determining the role of prothrombin in clot formation has been described by Quick.[256] This investigator frequently has demonstrated the production of hemorrhagic disease in chicks on diets deficient in vitamin K; the bleeding is apparently related to a prothrombin deficiency and occurs when the level of prothrombin is less than 20 per cent of normal. In human beings, bleeding usually occurs when there is a 90 per cent deficiency or more. Prothrombin deficiency may be produced not only by a deficient intake of vitamin K, as pointed out by Quick, but also by hepatic damage, failure of absorption and a direct toxic effect, as seen in sweet clover disease. Thayer and his colleagues,[257] as well as Ansbacher, [258] have also demonstrated the presence of a hemorrhagic tendency in chicks on deficient diets. They have been able to alleviate the bleeding by giving vitamin K. Ansbacher has recommended this method of assaying the potency of the product. Both the amount of material necessary and the duration of the therapeutic effect of the vitamin may be accurately determined.

A bleeding tendency and a prothrombin deficiency have been produced in dogs with biliary fistulas by Smith and his collaborators.[259] These authors conclusively demonstrated the failure of absorption of vitamin K when bile was absent from the gastrointestinal tract. The addition of bile to the normal diet restored the lowered prothrombin level to normal. The addition of vitamin K alone to the diet was of little value, as was also true of vitamins A and D. Similar clinical observations were

---

255. Almquist, H. J.; Pentler, C. F., and Mecchi, E.: Synthesis of the Antihemorrhagic Vitamin by Bacteria, Proc. Soc. Exper. Biol. & Med. **38**:336, 1938.

256. Quick, A. J.: The Nature of the Bleeding in Jaundice, J. A. M. A. **110**: 1658 (May 14) 1938.

257. Thayer, S. A.; McKee, R. W.; MacCorquodale, D. W., and Doisy, E. A.: Recovery from the Anemia Caused by a Diet Deficient in Vitamin K, Proc. Soc. Exper. Biol. & Med. **37**:417, 1937.

258. Ansbacher, S.: New Observations on the Vitamin K Deficiency of the Chick, Science **88**:221, 1938.

259. Smith, H. P.; Warner, E. D.; Brinkhous, K. M., and Seegers, W. H.: Bleeding Tendency and Prothrombin Deficiency in Biliary Fistula Dogs: Effect of Feeding Bile and Vitamin K, J. Exper. Med. **67**:911, 1938.

reported by Snell;[260] Butt, Snell and Osterberg,[261] and Warner, Brinkhous and Smith [262] for patients with obstructive jaundice. Those with a bleeding tendency associated with obstructive jaundice had a markedly reduced prothrombin level, which could be restored to normal by the addition of bile alone to the diet, or bile plus vitamin K, with the subsequent elimination of the hemorrhages. Dam and Glavind [263] stated that thrombopenia, aplastic anemia, hemophilia, nontropical sprue, myeloma, acute hepatitis, obstructive jaundice, cholelithiasis and carcinoma have a definite influence on the clotting time. It is their impression also that the bleeding tendency is a result of vitamin K deficiency, with an associated decrease in the prothrombin level.

Ferguson [264] stressed the importance of the quantitative relations of calcium and cephalin in thrombin formation. By means of a carefully standardized and controlled technic the author has demonstrated that calcium is a more fundamental determinant of the thrombin formation and that cephalin has a secondary influence up to a certain point in increasing the thrombin formation. Quick [265] has pointed out that antithrombin is an albumin or a substance closely associated with this fraction. Heparin is not antithrombin, but because of its affinity for the albumin, it intensifies the inherent antithrombic activity of this material. MacFarlane [266] has reported a case of fibrinopenia in a young boy with an associated bleeding tendency.

The role of the blood platelets in hookworm anemia has been studied by Landsberg.[267] He produced the disease by infecting dogs, but regard-

260. Snell, A. M.: Symposium on Hemorrhagic Diathesis in Cases of Jaundice: Its Relation to Vitamin K Deficiency; Preliminary Report; Clinical and Experimental Conditions Associated with Deficiency of Prothrombin, Proc. Staff Meet., Mayo Clin. **13**:65, 1938.

261. Butt, H. R.; Snell, A. M., and Osterberg, A. E.: The Use of Vitamin K and Bile in Treatment of the Hemorrhagic Diathesis in Cases of Jaundice, Proc. Staff Meet., Mayo Clin. **13**:74, 1938. Snell, A. M.; Butt, H. R., and Osterberg, A. E.: Treatment of the Hemorrhagic Tendency in Jaundice, with Special Reference to Vitamin K, Am. J. Digest. Dis. **5**:590, 1938.

262. Warner, E. D.; Brinkhous, K. M., and Smith, H. P.: Bleeding Tendency of Obstructive Jaundice: Prothrombin Deficiency and Dietary Factors, Proc. Soc. Exper. Biol. & Med. **37**:628, 1938. Brinkhous, K. M.; Smith, H. P., and Warner, E. D.: Prothrombin Deficiency and the Bleeding Tendency in Obstructive Jaundice and in Biliary Fistula: Effect of Feeding Bile and Alfalfa (Vitamin K), Am. J. M. Sc. **196**:50, 1938.

263. Dam, H., and Glavind, J.: Vitamin K in Human Pathology, Lancet **1**: 720, 1938.

264. Ferguson, J. H.: Quantitative Relationships of Calcium and Cephalin in Experimental Thrombin Formation, Am. J. Physiol. **123**:341, 1938.

265. Quick, A. J.: The Normal Antithrombin of Blood and Its Relation to Heparin, Am. J. Physiol. **123**:712, 1938.

266. MacFarlane, R. G.: A Boy with No Fibrinogen, Lancet **1**:309, 1938.

267. Landsberg, J. W.: The Blood Platelets in Hookworm Anemia, Am. J. Hyg. **27**:316, 1938.

less of whether or not the disease was fatal, there were no alterations in the circulating platelets or any noticeable toxic effect on the bone marrow.

BANTI'S DISEASE

Since 1881, when Banti described the disease which bears his name, sufficient evidence has accumulated to question the existence of this syndrome. Banti postulated that an unidentified toxic agent is the etiologic factor and that this substance is carried to the spleen, with secondary involvement of the liver. Sclerosis of the splenic vessels, atrophy of the malpighian corpuscles, induration of the pulp, sclerosis of the portal system and atrophic cirrhosis of the liver were the pathologic changes observed. Splenectomy effects a cure.

Menon [268] produced splenomegaly in rats and rabbits by narrowing the lumen of the portal vein for various lengths of time. The splenic sinuses appeared dilated, and there were distention of the pulp and trabecular veins, variable atrophy of the pulp and only a slight fibrillary increase. Hyperplastic reactions were absent. Complete obstruction of the portal vein was invariably fatal to the rats in three to forty-eight hours and to the rabbits in twenty to ninety-six hours. He concluded that portal obstruction is probably not the cause of Banti's disease. However, Hines and Fitzgerald [269] offered evidence from the autopsy study of a patient with so-called Banti's disease showing that the maintenance of hypertension in the sclerotic portal system is the etiologic factor in the production of the pathologic changes characteristic of hepatolienal cirrhosis.

Kemp [270] and Howells [271] have reviewed reports of large series of cases of Banti's disease. The outstanding symptoms and signs, in the order of their frequency, are splenomegaly, weakness, hemorrhage, enlargement of the liver, jaundice and ascites. The anemia present may be microcytic or macrocytic. The former is undoubtedly due to acute or chronic hemorrhage. Howells divided his cases into three groups according to the severity of the anemia and stated that approximately 81 per cent of the patients with severe anemia had hematemesis, whereas in the group with slight anemia, no hemorrhage was observed. The macrocytic anemia is probably due to the interference of erythropoiesis as a result of failure of the abnormal liver to store or present for

268. Menon, T. B.: Venous Splenomegaly: A Study in Experimental Portal Congestion, J. Path. & Bact. **46**:357, 1938.

269. Hines, L. E., and Fitzgerald, B.: Splenomegaly of the Banti type: Report of a Case with Postmortem Observations Four Years After Splenectomy, Arch. Path. **26**:155 (July) 1938.

270. Kemp, R.: Recurrent Haematemesis in "Banti's Disease," Brit. M. J. **1**: 222, 1938.

271. Howells, L.: Treatment of Splenic Anaemia and Banti's Syndrome, Lancet **1**:1320, 1938.

utilization to the bone marrow the substance necessary for the maturation of red blood cells.

Although there is considerable disagreement as to the existence of this disease, Rubnitz,[272] Alessandri [273] and De Candia and Floriani [274] stressed the importance of an accurate diagnosis in governing the selection of proper treatment. De Candia and Floriani suggested the intramuscular use of thymic extract as a method to differentiate the splenomegalies. They could produce an increase in the number of red blood cells, leukocytosis and splenic contraction in normal persons, as well as those with malaria, by intramuscular injections of thymic extract, but no changes were observed in patients with Banti's disease; hence, the suggestion that this test could be employed as a diagnostic procedure.

Splenectomy has long been considered the accepted treatment for this type of splenomegaly. A résumé of the early literature is misleading, as many of the reported cases were not cases of Banti's disease. Further, the disease may run a protracted course, so that any conclusions concerning the benefits derived from splenectomy must be guarded. Alessandri stated that removal of the spleen is indicated if portal thrombosis is not present. Dods [275] believes that operation is to be recommended only in the early course of the disease but is not enthusiastic about the results. Kemp [270] has suggested ligation of the varices as well as splenectomy or ligation of the splenic artery. Howells [271] is of the opinion that splenectomy does not affect the disease process. Iron may or may not be of value in eliminating the anemia. In his series, patients treated conservatively on a medical regimen lived as long as or longer than those with the spleen removed. He has suggested that the apparent difference in the life span of those operated on and of those not operated on may be due to the high mortality rate associated with the surgical procedure.

INFECTIOUS MONONUCLEOSIS

In a most comprehensive review Tidy [276] has summarized the various features relative to glandular fever and infectious mononucleosis. The disease was first described by Filatow, in 1885. Pfeiffer, in 1889; Desplats and Horschelmann, in 1894; Sprunt and Evans, in 1920; Lancope, in 1920, and Downey and McKinley, in 1923, have all made important contributions relative to this disease.

272. Rubnitz, A. S.: Blood Changes Associated with Different Splenomegalies, Nebraska M. J. **23**:87, 1938.

273. Alessandri, R.: Experiences with Surgery of the Spleen: Report of Two Unusual Cases, J. Mt. Sinai Hosp. **4**:489, 1938.

274. de Candia, S., and Floriani, F.: Azione del timo sull'emopoiesi e sulla milza, Policlinico (sez. prat.) **45**:1553, 1938.

275. Dods, L.: Indications for Splenectomy in Paediatric Practice, M. J. Australia **1**:377, 1938.

276. Tidy, H. L.: Glandular Fever and Infectious Mononucleosis, St. Thomas's Hosp. Rep. **2**:104, 1937.

As pointed out by Tidy, various organisms have been isolated and described as the etiologic factor of infectious mononucleosis, but none of them has ever been confirmed by any one other than the original investigator. The disease has occurred in various parts of the world in sporadic or epidemic form. Persons of either sex or of any age may be afflicted with glandular fever, though it is much more common in persons under 40, especially children.

The clinical course of the disease, as emphasized by Blum,[277] may be divided into three types: glandular, involving the lymphoid tissue in the cervical, axillary and inguinal regions; angiose, in which the symptoms involving the throat are outstanding; and the febrile type, which is most difficult to diagnose. The most common symptoms, as described by various clinicians, are general malaise, headache, tenderness of the glands, sore throat, chilliness, backache, coryza, anorexia, sweating, weakness, cough, dizziness, soreness and bleeding of the gums, nausea, stiff neck, epistaxis, stomatitis, abdominal pain, rash, photophobia and conjunctivitis. Trible [278] commented on the oral lesions occurring in the various blood dyscrasias. It is his opinion that infection is the primary cause and that the resulting blood picture enhances the condition. Beliveau and Russell [279] described a case of infectious mononucleosis in a 19 year old girl in whom the outstanding symptoms were similar to those of acute appendicitis. These authors also emphasized the symptoms related to the throat and stated that they may occur late in the course of the malady.

The more common physical signs are fever, enlargement and tenderness of the glands, injection and ulceration of the mouth and throat, enlargement of the spleen, hypertrophy of the tonsils and a maculopapular rash. Although enlarged glands are present in practically all cases and tenderness in more than half, suppuration of the glands is most rare.

The prognosis is excellent. The disease is apparently self limited, with relapses and remissions not uncommon. Often the course may be protracted, and both glandular and hemopoietic changes may be present for months.

The diagnosis usually can be made by the characteristic clinical and laboratory features. Leukocytosis of varying degree is present at some time during the course; leukopenia may be observed at the onset. There is present in increased numbers an atypical mononuclear cell, which is generally considered to be a lymphocyte, though all investigators will

277. Blum, L. L.: Infectious Mononucleosis, J. Indiana M. A. **31**:296, 1938.

278. Trible, G. B.: Clinical Manifestations in the Nose, Throat and Sinuses in Diseases of the Mononucleosis Type, Tr. Am. Acad. Ophth. **42**:367, 1937.

279. Beliveau, R. A., and Russell, B. W.: Infectious Mononucleosis (Glandular Fever): Report of a Case Presenting Symptoms of Acute Appendicitis, Maine M. J. **29**:167, 1938.

not concur with this opinion. The heterophile antibody test (Paul and Bunnell) appears to be practically specific for glandular fever (serum sickness to be excluded). Davidsohn [280] has again confirmed the value of this procedure and has devised a presumptive test which he believes will be of greatest value in borderline cases. The nature of the antibodies in infectious mononucleosis has received considerable attention. Bernstein [281] and Hatz [282] have called attention to the falsely positive reaction to the Wassermann test and its occurrence in this disease. These authors are of the opinion that the antibodies for these tests are not the same. Davidsohn [283] studied the isoagglutinins in various leukopoietic disorders and observed that the titers in infectious mononucleosis are within the normal range.

Since the disease is self limited and the cause unknown, no specific treatment has been devised. Rest in bed, forcing of fluids, a high caloric diet and any other necessary palliative measures are indicated.

GRANULOCYTOPENIA

Since the widespread recognition of granulocytopenia, following the clinical description of the syndrome by Schultz in 1922, many articles concerning its etiology, pathology, prognosis and treatment have appeared. Reports relating to the disease during the past year have been concerned chiefly with three of its aspects, as follows: the etiologic significance of certain drugs, especially sulfanilamide; the apparent decreasing incidence of the disease, and the evaluation of various types of therapy.

An excellent summary of contemporary knowledge concerning the neutropenic states has been given by Kracke.[284] He considers that until recently leukopenia has been regarded as secondary because it is usually in association with some other disease. The conditions which are stated to be characterized by neutropenia are measles, German measles, mumps, influenza of the respiratory tract, chronic malaria, Brucellosis, typhus fever, typhoid and overwhelming infectious processes due to pyogenic organisms. Furthermore, neutropenia is frequently observed in Banti's disease, some of the lymphogranulomas, glandular tuberculosis and many conditions in which the bone marrow is directly involved, such as

280. Davidsohn, I.: Test for Infectious Mononucleosis, Am. J. Clin. Path. (Tech. Supp.) **2**:56, 1938.

281. Bernstein, A.: False-Positive Wassermann Reactions in Infectious Mononucleosis, Am. J. M. Sc. **196**:79, 1938.

282. Hatz, B.: Wassermann Reaction in Infectious Mononucleosis, with Report of a Case with Unusual Clinical Features, Am. J. Clin. Path. **8**:39, 1938.

283. Davidsohn, I.: Isoagglutinin Titers in Serum Disease, in Leukemias, in Infectious Mononucleosis, and After Blood Transfusions, Am. J. Clin. Path. **8**:179, 1938.

284. Kracke, R. R.: The Neutropenic Diseases, Bull. New York Acad. Med. **14**:725, 1938.

multiple myeloma, miliary tuberculosis and bone marrow carcinomatosis. Although it is stated frequently that secondary neutropenic states are brought about by the action of bacteria or their toxins, he considers that there is scant proof available in support of this. He is quite ready to admit, however, that in some cases there is an infectious factor. The explanation of the neutropenia in some of the infectious diseases, he believes, is that the available circulating leukocytes are drawn to the site of the infection in order to combat it. Benzene, gasoline fumes, excessive radiation from radium, roentgen rays and sunlight, dietary deficiency and adrenal insufficiency are also mentioned as causes of neutropenia.

A more extensive discussion has been given by Kracke of the etiologic significance of the pain-relieving drugs, especially aminopyrine, in relation to the syndrome of granulocytopenia. Although there are a number of reasons for considering that this substance is the cause of the syndrome in many cases, he considers that definite proof has come from two lines of research. The most conclusive of these is that neutropenia can be reproduced by the administration of as little as a single dose to a susceptible person. The other is the complete disappearance of the disorder from Denmark when the use of aminopyrine was forbidden by governmental decree. It is his opinion that arsphenamine, in addition to causing aplastic anemia, may be responsible likewise for granulocytopenia, in which the involvement of the marrow is limited exclusively to depression of the granulocytes. He likewise believes that dinitrophenol can produce marked leukopenia; but as the other severe untoward effects of the drug are now recognized, it is rarely used and is therefore of little importance in the causation of granulocytopenia.

Sulfanilamide, in the opinion of Kracke, can cause granulocytopenia and, with the increasing use of this drug, it is predicted that an increasing number of cases will be encountered. He was able to collect reports of 14 cases of the syndrome following its use. Usually leukopenia followed the administration of large doses over a considerable period. He was unable to discover instances in which the syndrome followed the administration of relatively small quantities of the drug, which led him to believe that the leukopoietic depression differs from that produced by aminopyrine. It is thought that sulfanilamide can at times cause increased activity of the leukopoietic centers which results in an extremely high leukocyte count, but no explanation has been offered for this apparent stimulation. In our opinion the occurrence of striking leukocytosis in a patient with an infection, especially a child, who receives the drug may be due entirely to the normal response of the bone marrow to the infection.

In the aforementioned article a discussion of the mechanism of the production of the disease has been included. It is believed that the initial change is cessation of cellular output, although we think that a better term would be reduction rather than cessation; this results in a decrease

in the number of granulocytes in the circulating blood which may precede the appearance of symptoms by several days. The mechanism by which the offending agent operates to depress the marrow function is not known, but, regardless of the cause, it is considered that there is frequently an arrest of maturation, as first suggested by Fitz-Hugh and Krumbhaar. The clinical symptoms are coincident with bacterial invasion.

Two important diagnostic points are emphasized by Kracke. The first is that the patient with granulocytopenia seldom shows any disturbance of the hemoglobin, red blood cells or platelets of the circulating blood unless the process is prolonged. It is made clear, however, that there is no reason why a patient with granulocytopenia should not have associated anemia. This is especially true of those who have previously been suffering from other diseases. We should like to insist, however, that the presence of severe associated anemia often casts considerable doubt on the diagnosis of true granulocytopenia. A second point about which there appears to be general agreement is that these patients may exhibit a marked degree of prostration which seems entirely out of proportion to the physical signs. In some instances, the physical examination may yield no other information except evidence of fever and prostration. In our opinion, it is also true that there may be definite leukopenia at a time when few, if any, symptoms are present.

Kracke regards the present method of treatment as highly unsatisfactory and considers that "no one man probably has treated a sufficient number of cases to form a proper evaluation of therapeutic agents." He has pointed out that a patient with granulocytopenia is suffering from two distinct diseases; "not only is the patient depleted of granulocytes, but in addition to this, often times has become overwhelmed with invading organisms." On the basis that granulocytopenia is an emergency illness and that the use of any therapeutic agent of even possible value is justifiable, it is his practice to give repeated blood transfusions, liver extract in "ten times the ordinary dosage by injection," pentnucleotide, yellow bone marrow by mouth and symptomatic therapy. Even with the use of these, he has admitted that the mortality rate in his recent cases very closely approximated that of the earlier years. We are not entirely in agreement with these conclusions regarding therapy. In the first place, owing to the decreasing incidence of granulocytopenia, it has not been possible to evaluate the newer therapeutic agents properly. Secondly, the mortality rate for those patients who have been treated symptomatically and who, it can positively be stated, have not been given aminopyrine in some form or another during the acute illness, is not known. We are in accord with his statement that much more can be done by preventing the syndrome than by treatment after it has once developed.

In a second comprehensive article Kracke [285] has evaluated the data accumulated since 1931 which deal with the etiologic relation of certain drugs to granulocytopenia. The evidence is summarized regarding the etiologic significance of aminopyrine, arsphenamine, dinitrophenol and sulfanilamide. It is considered that conclusive proof is available which indicates that the disease may be caused by aminopyrine. This occurs only in an occasional person of the many who take the drug. Furthermore, it appears reasonable to conclude that the disease appears only in those who are allergic or sensitive to aminopyrine; therefore, the quantity which is administered bears little relation to the production of the disease. Incriminating information which points to the etiologic role of the substance is as follows: First, the disease occurs most frequently in those parts of the world where aminopyrine has the greatest use. Studies of the sharply declining incidence in Denmark, where the drug is now prohibited by law, are especially convincing. Second, its occurrence is greatest among those persons in any given vicinity who are the greatest users of aminopyrine. Third, a large majority of patients who have granulocytopenia are known to have taken the drug a short time prior to the onset of the condition. Fourth, and this is the most convincing evidence, the administration of the drug to patients who have recovered from the disease is followed by a reduction in the number of polymorphonuclear leukocytes in the peripheral blood. It has been emphasized that granulocytopenia may follow the use of a drug closely allied to aminopyrine, which is marketed under the name of novaldin (sodium phenyldimethylpyrazolon methylaminomethane sulfonate). It has the same chemical formula except that one of the methyl groups attached to the amino nitrogen is replaced by the sodium salt of methylsulfonic acid. The etiologic significance of this preparation has been demonstrated by the occurrence of leukopenia after its administration to a patient who had suffered previously from an attack of granulocytopenia induced by aminopyrine. It has been mentioned that antipyrine, which is the chemical precursor of and closely related to aminopyrine, has been suspected of producing the disease, but it appears to us that convincing evidence in this regard is lacking.

Kracke stated that the frequency of the disease in the southeastern part of the United States has decreased, as "the condition is now rarely seen," and concluded that this is also true throughout the country. He cited information, however, supplied by the United States Bureau of Vital Statistics for the years 1931 to 1936, inclusive, which indicates that there has been little change in the occurrence of the disease, as recorded in the mortality statistics, during these six years. These data should be accepted with the reservation, however, that errors of diagnosis might make them inexact. It is not inconceivable that other disorders,

285. Kracke, R. R.: Relation of Drug Therapy to Neutropenic States, J. A. M. A. **111**:1255 (Oct. 1) 1938.

such as aplastic anemia or overwhelming sepsis, might be regarded incorrectly as true granulocytopenia. On the other hand, improved methods of diagnosis might lead to the discovery of more cases, and as a result of a better understanding of the condition, it is possible that recovery occurs more commonly than in previous years. In our opinion, there has been a decrease in the incidence of the disease in recent years throughout the United States. There is no convincing information available, however, which indicates conclusively that this is due to the less frequent use of aminopyrine or allied substances either alone or in combination with other drugs. It can be suspected that this is the case because reliable data exist which indicate that the decrease of the incidence of the disease in Denmark paralleled the less frequent use of the drug.

It is contemplated by Kracke that granulocytopenia will continue to follow the administration of aminopyrine for two reasons: First, an occasional physician will continue to prescribe it; second, it will be used unknowingly in combinations with other pharmaceutic agents. Physicians are advised to refrain from the use of aminopyrine or analgesic agents that contain it, and patients should be warned against the dangers of self medication, particularly the purchase and use of pain-relieving remedies for which the formulas are not available. In general, we consider that this advice is sound, but certainly in one disease, acute rheumatic fever, it is the drug of choice for those who cannot tolerate the salicylates. The judicious use of aminopyrine in this condition cannot be condemned.

Other drugs which have been reported as possibly causing granulocytopenia have been discussed by Kracke. He concluded that arsphenamine, dinitrophenol and sulfanilamide are also unquestionably capable of producing the disease. Others which have been suspected by various writers are novaldin (a derivative of aminopyrine), antipyrine, acetophenetidin, acetanilid, cinchophen, neostibosan (an antimony compound), quinine and gold salts. A fairly complete list of the trade names of various preparations containing aminopyrine has been recorded in his article and should be useful to every physician.

It is entirely possible that all the drugs mentioned by Kracke and perhaps others as yet unrecognized are capable of producing the syndrome of granulocytopenia. Before accepting the etiologic relation of any given drug to the disease, however, the following points must be considered: First, has it been demonstrated that the drug can produce definite leukopenia in a patient who has recovered from the disease which it is alleged to have caused? Second, is there reasonable proof that other recognized leukocyte-depressing drugs have not been taken? Third, can it be established that the leukopenia is a true granulocytopenia and not due to sepsis or associated with some other blood dyscrasia? Until

these criteria have been satisfied, the evidence in regard to any given drug cannot be regarded as unequivocal.

Kracke has referred to the review by Plum and the report by Blew dealing with the etiologic relation between arsphenamine, with and without associated bismuth therapy, and granulocytopenia. In this summary 14 cases are reported in which the hematologic defect was limited to granulocytopenia, and there was no anemia or hemorrhagic diathesis. Other reports confirm this. It has long been known that arsenic compounds can produce aplastic anemia, but it is important to establish that they can also cause true granulocytopenia. The evidence is suggestive but not conclusive that this is the case.

Kracke has summarized the data regarding 11 cases of granulocytopenia following the administration of sulfanilamide which have been reported since April 1937 and has added 2 of his own. Death occurred in 10 of the 13 cases. In his opinion, this drug should be added to those which are capable of producing the disease. As the syndrome usually developed after the administration of large amounts of the drug (40 to 50 Gm.), he has reemphasized that the mechanism of granulopoietic depression may be different from that following the administration of a single dose of aminopyrine. We wish to stress, however, that unless an idiosyncrasy exists, it is difficult to understand why in only a very rare patient among the many thousands who have received this drug in the past few years the condition develops.

Fitz-Hugh [286] stated that agranulocytic angina or, as he called it, pernicious leukopenia is now almost universally believed to be caused by an acquired sensitivity of the leukocytes, endothelium and leukopoietic tissues to certain drugs and other allergens (or potential toxins). He regards sensitivity, hypersensitivity and allergy as synonymous terms and, along with the expression idiosyncrasy, as meaning drug allergy. When the latter term is used, he considers that there has been a prior specific sensitizing contact, probably acting through a drug-protein combination, which is followed by an allergic response on repetition of contact with the drug. This reaction may be precipitated by a minute dose of the substance, and it does not necessarily resemble the usual pharmacologic or toxic action of the drug. He listed the drugs which have been suspected or proved to be the allergic cause of granulocytopenia as follows: "aminopyrine, dinitrophenol, arsphenamine and neoarsphenamine, gold salts, the diethylamine salt of stibanilic acid (neostibosan), acetophenetidin, sodium phenyldimethyl pyrazolon methylaminomethane sulfonate (novaldin), a preparation containing about equal molecular parts of aminopyrine and 8-hydroxyquinoline-5-sulfonic acid [causalin], quinine, cinchophen, bismuth, 5,phenyl-5,ethyl-

286. Fitz-Hugh, T., Jr.: Sensitivity Reactions of the Blood and Bone Marrow to Certain Drugs, J. A. M. A. **111**:1643 (Oct. 29) 1938.

hydantoin (nirvanol), plasmoquine 8-dimethylamino-iso-amyl 6 methoxyquinoline (plasmochin) and sulfanilamide." Other causes which Fitz-Hugh would classify as "possible conditioning factors" in drug allergy are as follows: endocrine disturbances, bacterial sensitization, cyclic toxemia, fatigue states, accidental and surgical trauma, genetic predisposition and vitamin deficiency. It is obvious that such an amazing group of possibilities can mean only that these conditioning factors, if they are of importance, have not been established on a firm scientific foundation.

It is emphasized again by this author that the mechanism leading to agranulocytosis is to be found in the arrest of maturation of the leukocytes in the bone marrow. We believe that this is the most acceptable idea of the pathologic change in the disease. We see no reason, however, why the phenomenon of "sticking" of leukocytes to vascular endothelium should be postulated to explain the sudden temporary disappearance of granulocytes from the circulation (granulocytoclastic crisis).

Fitz-Hugh has considered all the various forms of therapy in the treatment of this condition and emphasized that yellow bone marrow concentrate for oral administration is of real value.

The Relation of Sulfanilamide to Granulocytopenia.—Since sulfanilamide and its derivatives and allied substances are now commonly used in the treatment of various types of bacterial infection, it becomes increasingly important to consider the etiologic relation of these drugs to the syndrome of granulocytopenia. In 1937 there were 8 and in 1938 there were 9 published articles bearing on this subject.

Schwartz, Garvin and Koletsky [287] have reported a case of granulocytopenia which developed after the administration of 48.6 Gm. of sulfanilamide over a period of nineteen days. The leukocyte count fell to 800 per cubic millimeter, and no granulocytes were present. The lowest recorded level of the erythrocytes was 3,410,000 per cubic millimeter. Pentnucleotide (10 cc. twice daily), liver (5 cc. twice daily) administered intramuscularly and a transfusion of 500 cc. of whole blood and typhoid vaccine intravenously were given without effect. The patient had a primary syphilitic lesion (probably a reinfection) and had received antisyphilitic therapy. The last dose of neoarsphenamine had been given three years previously; mepharsen was last given fifty-six days before the development of leukopenia; bismuth and mercury had been administered intermittently for more than five years without signs of intolerance. Necropsy showed a maturation arrest of the myeloid series. There was marked phagocytosis of iron in the liver, spleen and bone marrow, which was interpreted as evidence that the anemia was hemolytic.

287. Schwartz, W. F.; Garvin, C. F., and Koletsky, S.: Fatal Granulocytopenia from Sulfanilamide, J. A. M. A. **110**:368 (Jan. 29) 1938.

The case of a man aged 22 who was treated with sulfanilamide for acute gonorrhea was reported by Berg and Holtzman.[288] The drug was administered in 10 grain (0.65 Gm.) doses every four hours. After five days the use of this medication was discontinued on account of slight fever, cramps, gaseous eructations and nausea. After an interval of four days, it was resumed, with the dose of 5 grains (0.32 Gm.) three times daily. Seven days later there was a recurrence of the same toxic symptoms, and the use of the medication was again discontinued. It was resumed once more, with a dose of 10 grains thrice daily, despite evidences of toxicity. After eleven days the patient experienced chill and fever (102 F.), and nausea, vomiting and prostration developed. The lowest leukocyte count recorded was 1,600 per cubic millimeter (all lymphocytes); the erythrocyte count fell to 2,600,000 per cubic millimeter and the hemoglobin value to 60 per cent. We wish to emphasize that this case illustrates the need for the warning which is now well recognized, viz., that if the serious toxic manifestations of the drug, such as nausea, vomiting and fever, occur, they are likely to reappear if use of the drug is resumed after an interval.

Long, Bliss and Feinstone [289] reported on a patient with gonorrheal urethritis and arthritis who manifested the typical picture of agranulocytic angina toward the end of the third week of treatment with sulfanilamide. Procedures designed to rid him of the sulfanilamide were immediately instituted, and recovery followed within ten days. These observers recorded the frequency of granulocytopenia as 0.3 per cent in their group of 307 adults and 101 children receiving sulfanilamide therapy.

Jones and Miller [290] reported the case of a man aged 26 who was given 2.4 Gm. of sulfanilamide daily for ten days for acute gonorrhea. At the end of this time he had headache, backache and fever (104.4 F.); the leukocyte count was between 9,000 and 10,000 per cubic millimeter, and there was moderate injection of the pharynx. The use of the drug was discontinued, and the symptoms subsided with the fever; sulfanilamide therapy was resumed, with doses of 0.9 Gm. daily, being increased by that amount each day until a daily dose of 3.6 Gm. was reached. At this time the temperature again rose, and he had the feeling that "he was coming down with a cold." The leukocyte count remained normal. Therapy was again stopped, and the symptoms disappeared. After an

288. Berg, S., and Holtzman, M.: Fatal Granulocytopenia Following Sulfanilamide Therapy, J. A. M. A. **110**:370 (Jan. 29) 1938.

289. Long, P. H.; Bliss, E., and Feinstone, W. H.: Mode of Action, Clinical Use and Toxic Manifestations of Sulfanilamide, J. A. M. A. **112**:115 (Jan. 14) 1939. Long, P. H., and Bliss, E. A.: Toxic Manifestations of Sulphanilamide, Ann. Surg. **108**:808, 1938.

290. Jones, H. W., and Miller, C. P.: Neutropenia Following Sulfanilamide: Report of a Case, J. Lab. & Clin. Med. **24**:121, 1938.

interval of four days, 2.7 Gm. of the drug was administered on one day only. The next morning the leukocyte count was 4,100 per cubic millimeter, with 25 per cent granulocytes. The patient complained of headache, chilliness and general malaise. On the following day the leukocyte count was 2,300, with 5 per cent neutrophils. Yellow bone marrow concentrate, 80 drops daily, was given, after which the leukocyte count rose rapidly. Four days later he was symptomless. It is interesting to note that the sulfanilamide content of the blood was 5.6 and 2.8 mg. per hundred cubic centimeters during the second and third periods of intoxication, indicating that an overdosage, in the ordinary meaning of the term, had not been given. The authors concluded that the patient had acquired an increased susceptibility to the drug. The course of events in this case, we believe, illustrates clearly that a febrile reaction which develops during the course of sulfanilamide therapy is a significant warning which cannot be disregarded.

Johnston [291] reported on 2 patients with granulocytopenia following the use of sulfanilamide whom he had observed and 10 previously reported on in the literature. These were studied in an attempt to discover common features which might help in the prevention, recognition and treatment of this condition. The first case observed by him was that of a woman of 23 years with severe puerperal infection due to Streptococcus haemolyticus who was given 61.3 Gm. of neoprontosil and sulfanilamide orally and intramuscularly over a period of twenty days. On the eighteenth day of treatment the leukocyte count was found to be 1,520 per cubic millimeter, and no polymorphonuclear neutrophilic cells were present in the blood film. "Marked secondary anemia" was also present. Recovery followed discontinuance of the use of sulfanilamide and prontosil, and the administration of pentnucleotide and a blood transfusion. It is important to record that during her illness, the patient received 10 tablets of allonal (allylisopropylbarbituric acid with aminopyrine), each of which contained 0.03 Gm. of aminopyrine, over a period of twenty-one days. The second case was that of a woman aged 33 years with moderately severe puerperal infection due to Str. haemolyticus who had been treated with prontosil orally and intramuscularly for three days prior to admission to the hospital. She received a total of 68.45 Gm. of sulfanilamide and neoprontosil over a period of twenty-two days. The leukocyte count fell to 3,500 per cubic millimeter, with 41 per cent of the cells of the polymorphonuclear neutrophil series. There was also anemia, as indicated by a hemoglobin value of 56 per cent and a red blood cell count of 2,600,000 per cubic millimeter. She recovered after discontinuance of treatment with sulfanilamide, the administration of pentnucleotide and a blood transfusion. It is noteworthy to record that this patient during her illness

291. Johnston, F. D.: Granulocytopenia Following Administration of Sulfanilamide Compounds, Lancet **2**:1044, 1938.

also received 5 tablets of allonal over a period of eight days. The observers were cognizant of the possible etiologic relation of allonal to the agranulocytosis in the 2 cases but stated that in these cases it was "not very clear."

Johnston recorded 10 additional cases of granulocytopenia which had been reported up to the time his article was written and tabulated the main details of each. Five more cases which have been less fully reported were also mentioned. He concluded that granulocytopenia with a high mortality rate may occur in certain susceptible persons after or during treatment with the sulfanilamide-prontosil group of drugs. It is not thought necessary to make routine leukocyte counts for all patients receiving these drugs, but frequent examinations of the blood should be made when there is pyrexia or an increase in pyrexia which is already present. It is his conclusion that the duration of treatment is probably a more important factor in relation to the disease than the total dosage of the drug.

McGuire and McGuire [292] reported the case of a woman aged 35 with rheumatoid arthritis who received 15 grains (0.97 Gm.) of sulfanilamide orally each day for thirty days and then received 5 cc. of prontosil (disodium 4-sulfamidophenyl-2′-azo-7′-acetylamino-1′hydroxynaphthalene-3′,6′-disulfonate) intramuscularly every six hours for three days. Ulceration of the throat and cervical adenopathy developed, and the leukocyte count was found to be 450 per cubic millimeter, with no granulocytes. As the patient denied taking any other drugs which were known to cause granulocytopenia, it was concluded that sulfanilamide may have been the exciting factor in this case. In an effort to prove this causal relation two months after recovery the patient was given 7.5 grains (0.49 Gm.) of sulfanilamide daily, and the blood was examined each day at the same time. After five days the total leukocyte count, which had been 13,050 per cubic millimeter, with 82 per cent granulocytes, had decreased to 9,250 per cubic millimeter, with 35 per cent granulocytes. The test was discontinued, although it was not recorded that the patient complained of symptoms. On the day after the use of the drug was stopped, the leukocyte count was 11,450 per cubic millimeter, with 62 per cent granulocytes. It was concluded that these observations were strong evidence that sulfanilamide was of importance in the production of the leukopenia. We believe that such studies as these are highly important and offer the only conclusive evidence that the drug can cause neutropenia, although significant supplementary data may be obtained by clinical studies. It is regrettable but understandable that more frequent leukocyte counts were not made, and the experiment continued until all question of the etiologic relation had been dispelled.

292. McGuire, P. R., and McGuire, J. P.: Agranulocytosis Following Use of Sulfanilamide: Report of a Case, Illinois M. J. **73**:425, 1938.

The case of an 11 year old girl with multiple articular involvement associated with fever, anorexia, emaciation and progressive anemia was reported by Alpert and Forbes.[293] After the first few days of sulfanilamide therapy there was leukocytosis (27,200 leukocytes, with 94 per cent granulocytes), which they were inclined to attribute to the medication. That this is not necessarily the case is indicated by a subsequent greater leukocytosis at a period when the drug had not been given for some time. A total dose of 12.2 Gm. was administered over a period of one week, and then the treatment was discontinued for a similar interval. At the end of that time, after 0.6 Gm. had been taken, the leukocyte count was found to be 3,200 per cubic millimeter, with a total disappearance of granulocytes from the circulating blood. The patient had a febrile reaction and vomited; oral lesions were present. The sulfanilamide treatment was discontinued. Convalescent streptococcus serum, three blood transfusions and four intramuscular injections of liver extract were given, and prompt improvement resulted, with subsequent complete recovery from the blood dyscrasia. No reference was made concerning the administration of other drugs of etiologic importance, such as aminopyrine, which must be considered as a possibility when the patient is suffering from an acute rheumatic condition.

Allen and Short[294] reported the development of agranulocytosis in an 18 year old girl during the course of treatment with sulfanilamide for a mild infection, presumably due to the gonococcus. There was spontaneous recovery without recourse to any of the recommended forms of therapy for neutropenia. In the authors' opinion, no other drugs of etiologic significance were administered, so that sulfanilamide could be considered, at least presumptively, as the cause of the granulocytosis. A total dose of 15.9 Gm. of the drug was given over a period of nine days without untoward effects. After its use was discontinued for eight days, it was resumed on account of the persistence of the vaginal discharge. Four hours after she received a dose of 1.0 Gm., fever developed; and after a second dose, the treatment was discontinued. Two days later it was resumed again, with a dosage of 2.4 Gm. daily for two days. At the end of this time the temperature reached 103 F., but there was no evidence of oral or pharyngeal sepsis. The leukocyte count was 2,900 per cubic millimeter, with 64 per cent polymorphonuclear neutrophils. The use of the drug was then permanently discontinued. The white blood cell count continued to decrease, and finally all polymorphonuclear neutrophils disappeared. The patient gradually improved and was discharged from the hospital twenty-four days after discontinuing the use of sulfanilamide. The

293. Alpert, G. R., and Forbes, R. P.: Granulocytopenia and Hyperleucocytosis Following Sulfanilamide Therapy, J. Pediat. **12**:605, 1938.

294. Allen, J. G., and Short, C. L.: Granulocytopenia Associated with Sulfanilamide Therapy, New England J. Med. **219**:6, 1938.

authors emphasized two points especially with reference to this case: In the first place, there was absolute monocytosis, which they believe is a good prognostic sign; second, the earliest objective sign of granulocytosis was the increase in body temperature. In their opinion, an idiosyncrasy rather than a cumulative toxicity is probably the mechanism in the production of granulocytopenia in these cases.

After a careful consideration of the literature, we believe that the following conclusions regarding the etiologic relation of sulfanilamide to granulocytopenia are warranted:

1. Considering the widespread use of sulfanilamide, there is an extremely small number of patients in whom the syndrome of granulocytopenia develops.

2. Sulfanilamide is usually given to patients with sepsis, which in itself may admittedly cause marked leukopenia.

3. To say that sulfanilamide causes granulocytopenia should mean that the significant alteration in the blood is limited exclusively to a decrease in the number of granulocytes.

4. In some cases in which granulocytopenia following sulfanilamide therapy has been reported, other drugs that are known to produce the syndrome have been administered at the same time. In some cases this information has been included in the report. In others, in which no mention has been made concerning the use of other drugs, it is conceivable that aminopyrine alone or in combination with other drugs may have been given.

5. No one has reported conclusive evidence that sulfanilamide will cause neutropenia when given to any one who has recovered from the syndrome of granulocytopenia.

6. In some instances, granulocytopenia has developed suddenly with the resumption of therapy after an interim of several days. This suggests that the initial dosage may have sensitized the patient and that the subsequent granulocytopenia might be considered properly as an allergic manifestation.

7. An increase in the height of the temperature when fever is already present or the development of fever in a patient who is receiving sulfanilamide should at once suggest the necessity for a careful examination of the blood to determine whether there is an alteration in the absolute number of polymorphonuclear leukocytes.

8. While pentnucleotide, blood transfusions, yellow bone marrow and liver extract may be of value in the treatment of patients who have granulocytopenia following sulfanilamide therapy, it is probable that the most important therapeutic procedure is the early and immediate discontinuance of the use of sulfanilamide.

Other Etiologic Factors.—Reznikoff [295] considers that there are four factors of importance in the causation of granulocytopenia. These are: (1) fatigue, (2) drugs, (3) menstruation and (4) infection. He found that in 11 of 13 cases in which information on this point was obtained, there was a history of fatigue antedating the onset of the granulocytopenia. We desire to emphasize that it is difficult to evaluate the etiologic significance of fatigue, because it may lead to the use of the very drugs which are in themselves known to cause granulocytopenia. Almost every patient in his series had taken aminopyrine, dinitrophenol, arsphenamine or allonal or had received roentgen treatment, which makes difficult the evaluation of fatigue as an etiologic possibility. If it is granted that fatigue may play a role in the causation of the disease other than that of leading to the use of drugs which might produce it, there is no explanation or theory offered as to how it might act. Although he agreed that drugs, especially aminopyrine, are important in causing granulocytopenia, he cited the figures of Jackson, who stated that in 44 per cent of his cases no drugs had ever been used; a statement which, in our opinion, is difficult to prove. Furthermore, the question is raised as to why a drug like aminopyrine may be taken for a considerable period of time without apparent ill effects and then the patient suddenly manifests the disease. He does not consider this in keeping with the present conception of allergy and has stated that if it is a sudden manifestation of an Arthus phenomenon, it must be admitted that the precipitating factor is unknown. This observer considers that in most cases infection is the result rather than the cause of the disease, but he still believes that it must be considered as one of the etiologic agents. Reznikoff believes that menstruation is undoubtedly an important etiologic factor in the disease and has stated that "many cyclical cases have been described in which this was the only etiologic condition and in which no drugs were known to have been used." Although it cannot be denied that menstruation may bear a relationship to the disease, the evidence at best is only presumptive. Here again, in appraising this matter, we believe that the situation is complicated because many women take aminopyrine alone or in association with other drugs for dysmenorrhea.

Reznikoff is in accord with the belief that monocytosis is a favorable prognostic sign in granulocytopenia. Of his 13 patients, 9 recovered, and all had sustained relative and in most instances absolute monocytosis, which in many cases preceded the return of the neutrophil count to normal, the rise in the leukocyte count and the drop in temperature or symptomatic improvement.

295. Reznikoff, P.: The Etiologic Importance of Fatigue and the Prognostic Significance of Monocytosis in Neutropenia (Agranulocytosis), Am. J. M. Sc. **195**:627, 1938.

Two cases of granulocytopenia are reported by Jackson [296] in which the etiologic agent appears to have been causalin, a drug made up of approximately equal parts of aminopyrine and hydroxyquinoline, and widely advertised for the treatment of "rheumatism." One patient, a woman aged 67, had been taking 3 tablets of causalin daily for a month. One week before admission to the hospital she had a severe sore throat and fever. The palate and fauces showed marked inflammatory change; the white blood cell count was 1,200 per cubic millimeter, with 4 per cent polymorphonuclear neutrophils. The red blood cell count was 3,300,000 per cubic millimeter and the hemoglobin value 59 per cent. Pentnucleotide, in doses of 40 cc. daily, failed to produce improvement, and the patient died sixty hours after entry. A second patient, a woman, had taken 200 tablets of causalin for the relief of arthritis in the three weeks prior to admission to the hospital. Eleven days after the initial dose of the drug was taken, fever and ulcerative lesions of the mouth developed. On entrance the leukocyte count was 1,600 per cubic millimeter; the only granulocytes present were basophils (2 per cent). The red blood cell count was 3,830,000 per cubic millimeter and the hemoglobin value 63 per cent. Recovery followed pentnucleotide therapy. The inherent dangers in the use of causalin, containing as it does both aminopyrine and a quinoline derivative, have been emphasized, and Jackson believes that there is no excuse for its therapeutic use.

Jackson [297] reported the case of a woman aged 37 in whom neutropenia developed, as evidenced by a leukocyte count of 1,500 per cubic millimeter, with 20 per cent granular leukocytes. This case is of interest for several reasons: First, the patient had consumed more than 10 gallons (38 liters) of elixir of phenobarbital in the five years prior to the onset of the granulocytopenia and, in addition, had taken an unknown quantity of phenobarbital tablets. Although the blood condition followed the excessive use of a hypnotic drug, there is no evidence that this drug was of etiologic significance in producing the condition. We are of the opinion that there is no convincing proof in the literature that hypnotic drugs alone, especially the barbiturates, will cause granulocytopenia, although many cases have been reported following the use of these drugs when combined with aminopyrine. In such cases it is generally accepted that the aminopyrine rather than the hypnotic drugs is responsible for the granulocytopenia. Furthermore, the possibility must always be considered that a patient who is so unstable as to take such large doses of phenobarbital frequently resorts to hypnotic drugs which may be combined with aminopyrine. Such combinations are readily purchasable in many states in this country without a prescription.

296. Jackson, H., Jr.: Agranulocytosis Following Ingestion of Causalin, J. A. M. A. **111**:523 (Aug. 6) 1938.

297. Jackson, W. C.: Case of Neutropenia Following Excessive Use of Phenobarbital, M. Rec. **148**:148, 1938.

Second, the case is of interest because there was associated anemia, as indicated by a minimum red blood cell count of 1,850,000 per cubic millimeter and a hemoglobin value of 32 per cent. The author emphasized, very properly, that it is unusual to observe such severe anemia in granulocytopenia and ascribed it to some complicating factor rather than a part of the acute disease. Recovery followed the use of blood transfusions, pentnucleotide and liver extract and the prohibition of hypnotic drugs.

In a man aged 46 years, reported on by Mays,[298] leukopenia, with chills and fever and jaundice developed after the administration of twenty-nine injections of arsphenamine, fourteen of neoarsphenamine and fifty-one of bismuth subsalicylate during a period of twenty months. Recovery followed five injections of pentnucleotide, infusions of dextrose and a high carbohydrate diet. The red blood cell count and the hemoglobin value remained within normal limits. In our opinion it is not possible to state definitely that the leukopenia was due to the antisyphilitic therapy; furthermore, the blood findings are somewhat puzzling and not entirely characteristic of granulocytopenia. On one occasion the leukocyte count was reported as 3,100 per cubic millimeter, "with apparently all very immature cells;" on another occasion the leukocyte count was 2,800 per cubic millimeter, with 2 per cent polymorphonuclear leukocytes, 4 per cent metamyelocytes and 28 per cent myelocytes.

Chapman [299] reported a fatal case of granulocytopenia, that of a woman aged 43 years, with fever, ulcerative lesions in the throat and a leukocyte count of 650 per cubic millimeter, with only 2 per cent polymorphonuclear cells. The red blood cell count was 3,780,000 per cubic millimeter and the hemoglobin value 70 per cent. The patient died, despite the use of pentnucleotide, parenteral treatment with liver extract and repeated small transfusions of whole blood. The cause of the condition was not discussed; the statement concerning drug therapy was that at the onset of the illness the patient took acetylsalicylic acid, a cathartic and a patent medicine called "H-C" powder, the chemical composition of which was not given.

Shaw [300] reported the case of a 64 year old woman with pyrexia; ulcerations of the mucous membranes of the posterior pharyngeal wall, tonsils and fauces, and a maximum reduction in the white blood cell count of 560 per cubic millimeter, with 17 per cent polymorphonuclear leukocytes. There was no statement which indicated positively that the

298. Mays, A. T.: Agranulocytosis Following Administration of Arsphenamines and Bismuth: Report of a Case, New York State J. Med. **38**:1234, 1938.

299. Chapman, W. H., Jr.: Agranulocytosis with Report of a Case, Virginia M. Monthly **65**:287, 1938.

300. Shaw, C. C.: Granulocytopenia: Report of a Case with Autopsy, New England J. Med. **218**:343, 1938.

patient did or did not receive certain drugs of etiologic importance, such as aminopyrine. This case was regarded as one of the group in which the condition can "develop in certain instances without any apparent cause." It is unusual, in our opinion that the ulcerations of the throat developed prior to a decrease in the leukocyte count.

Stites and Stites [301] observed a woman aged 32 who had fever, an injected throat and some enlargement of the anterior cervical lymph glands. The leukocyte count varied from 2,700 per cubic millimeter on admission to the hospital to 1,450 per cubic millimeter the fourth day afterward. Granulocytes were absent on the seventh day, and the count did not rise above 9 per cent for seven consecutive days. Recovery followed the giving of pentnucleotide and of liver extract parenterally and one blood transfusion of 520 cc. Six weeks before entry she had experienced chills and fever and was treated with quinine, atabrine and plasmochin. The authors implied that the atabrine and plasmochin were the cause of the granulocytopen Although this must be admitted as a possibility, we wish to emphasiz that there was no conclusive proof to substantiate this assumption. The increasing use of the new preparations in the treatment of malaria and their possible relation to granulocytopenia make this report noteworthy.

The case of a man aged 23, with dementia praecox who was found to have a leukocytic count of 400 per cubic millimeter, with 11 per cent granulocytes, 88 per cent lymphocytes and 1 per cent monocytes, was reported by Kitching.[302] The red blood cell count was 4,500,000 per cubic millimeter, a few normoblasts were present and an occasional cell showed punctate basophilia. No ulcerative lesions of the mucous membranes were apparent during life, but at necropsy the colon was the seat of extensive necrotic lesions which appeared to involve only the mucosa. The sternal bone marrow appeared normal to the naked eye. Smears showed absence of polymorphonuclear cells, but myelocytes and lymphocytes were present. The only drug which the patient had received was soluble barbital ("sodium barbitone"). The case is reported as one of primary granulocytopenia of unknown causation.

The case of a man aged 18 was reported by LaSalle and LaSalle.[303] He was in a coma when admitted to the hospital after three days of illness characterized by fever, sore throat and headache. There were 4,300 leukocytes per cubic millimeter, "of which only 2 per cent were granulocytes of the basophilic series." There were marked hypotension and redness of the throat but no other buccopharyngeal lesion. The patient succumbed one and a half hours after first being observed. No

301. Stites, J., and Stites, F. M.: Agranulocytosis, Kentucky M. J. **36**:324. 1938.

302. Kitching, E. H.: A Case of Agranulocytosis, Lancet **1**:83, 1938.

303. LaSalle, M., and LaSalle, C.: Fulminating Case of Agranulocytosis, M. Rec. **148**:9, 1938.

statement was made concerning the use of drugs which might have a bearing on the causation.

Of considerable etiologic importance may be the chance observation by Lawrence and Syverton [304] of a cat with only 350 white blood cells per cubic millimeter and no neutrophils in the peripheral blood. The supernatant fluid from a ground-up suspension of the liver of this animal was injected into 5 other cats, and the same blood picture developed in 2 of them. This process was continued until thirteen transmissions were accomplished. After a latent period of five days following the injection, in which the animals appeared normal, there were pyrexia, anorexia and listlessness; coincident with these changes there was marked neutropenia. There were no changes in the red blood cell count, and the platelet count remained normal or increased. After one or two days, at the height of the disease, the animals either succumbed or had an increase in the white blood cell count and recovered. The causative agent, in the opinion of these observers, appeared to be some filtrable agent or a virus.

Therapy.—The observation by Giffin and Watkins [305] that after the administration of yellow bone marrow to patients with "secondary anemia" there was an increase in the number of neutrophils and monocytes in the circulating blood led to the therapeutic use of this substance in cases of granulocytopenia. Twenty-four patients were treated, and 2 of those who received adequate therapy died. The authors believe that the therapeutic results obtained with bone marrow have been sufficiently satisfactory to warrant a continuation of its administration to the exclusion, at least temporarily, of other methods of treatment, such as treatment with liver extract, transfusion and pentnucleotide. A daily dose of 200 to 300 grains (13 to 20 Gm.) was recommended; after recovery a daily dosage of 50 to 100 grains (3.3 to 6.5 Gm.) was given three or four months or longer to prevent relapse. It is their belief that some substance in the bone marrow may have the power of stimulating the production or maturation of the leukocytes. They emphasized, very properly, that spontaneous recoveries, multiple methods of treatment and inaccurate diagnoses make difficult the accurate appraisal of various forms of therapy in this disease.

The studies of Marberg and Wiles [306] were inspired by the statement of Dr. C. H. Watkins, of the Mayo Clinic, at the meeting of the American Medical Association in 1933 that yellow bone marrow had been beneficial in cases of granulocytopenia. A nonsaponifiable fraction of yellow bone marrow was prepared and administered to 6 normal persons

304. Lawrence, J. S., and Syverton, J. T.: Spontaneous Agranulocytosis in the Cat, Proc. Soc. Exper. Biol. & Med. **38**:914, 1938.

305. Giffin, H. Z., and Watkins, C. H.: The Administration of Yellow Bone Marrow in Agranulocytic Angina, Minnesota Med. **21**:62, 1938.

306. Marberg, C. M., and Wiles, H. O.: Granulocytopoietic Fraction of Yellow Bone Marrow, Arch. Int. Med. **61**:408 (March) 1938.

without effect on the leukocyte counts. Seven patients with leukopenia which was not considered to be true granulocytopenia but was associated with such conditions as aplastic anemia and leukemia showed no response to the administration of the concentrate. Six patients with agranulocytic angina (malignant neutropenia) were treated with yellow bone marrow per os, and all recovered. The authors concluded that yellow bone marrow concentrate has granulocytopoietic activity sufficient to bring about a normal blood picture when used in cases of agranulocytic angina and of some other leukopenias. In our opinion this conclusion does not appear to be well grounded for the following reasons: 1. Spontaneous improvement, without relation to therapy, may well have explained why the patients recovered. 2. Recovery may well have been due to the discontinuance of the use of aminopyrine, which some of the patients were known to have received, 3. Only 1 of the patients received yellow bone marrow concentrate as the sole granulocytopoietic medication; the others were also given liver extract or pentnucleotide therapy in addition. In attempting to appraise different forms of therapy for acute agranulocytic angina, it should be kept in mind constantly that many patients will recover if the cause of the condition, which in the past has most frequently been aminopyrine, is removed.

Netousek [307] reported that in a case of aminopyrine granulocytopenia, transfusion with blood from a patient with myelogenous leukemia was followed by a good therapeutic result after pentnucleotide and a blood transfusion had failed to produce benefit.

Massias and Quat [308] observed recovery of a patient following the administration of vitamins A, B and C. The white blood cell count was 5,400 per cubic millimeter, and polymorphonuclear cells were absent from the circulating blood. The condition was attributed to bismuth therapy which had been used as an antisyphilitic measure. Two days after the vitamin therapy had been instituted the granulocyte count had increased to 31 per cent and in five days to 74 per cent.

HODGKIN'S DISEASE AND LYMPHOSARCOMA

Webster [309] noted periodicity in the recurrence or exacerbation of symptoms in a number of neoplastic conditions, including leukemia, lymphadenoma and Hodgkin's disease. Periods of increased growth came in cycles of thirty-three weeks, with a lesser peak at half periods (sixteen and one-half weeks). Half periods were most marked in Hodg-

307. Netousek, M.: Traitement de l'agranulocytose par le sang leucémique, Sang **12**:345, 1938.

308. Massias, C., and Phan Huy Quat: Guérison rapide d'un syndrome agranulocytaire post-bismuthique par la vitaminothérapie A, B, C, Sang **12**:363, 1938.

309. Webster, J. H. D.: Periodicity in Cancer and Other Neoplastic Diseases (Four Hundred and Fifty Cases), Brit. J. Surg. **26**:113, 1938.

kin's disease and sarcoma. The data have been interpreted to indicate a virus causation of these diseases.

Bonnet, Thieffry and Montefiore [310] isolated avian tubercle bacilli from Hodgkin's lymph nodes, while Donati [311] and Vasconcellos [312] found Rickettsia in their material.

In 5.5 per cent of 450 cases of Hodgkin's disease and lymphosarcoma, Decker, Leddy and Desjardins [313] found leukopenia (less than 5,500 leukocytes per cubic millimeter). From the point of view of roentgen therapy, leukopenia is not a contraindication to treatment, nor does it have prognostic significance. Initial leukocyte counts between 5,500 and 10,000 occurred in 58.5 per cent of the series and counts above 10,000 in 36 per cent. Polymorphonuclear percentages were higher, with lymphopenia in those cases in which there was an elevated leukocyte count. In the leukopenic normal and leukocytosis patients, the losses in percentage of leukocytes four to eight weeks after irradiation were, respectively, 16 and 9.9 per cent, 22.4 and 33.3 per cent and 58.6 and 50.9 per cent. In the last group the reduction was due primarily to a decrease in the polymorphonuclear level rather than a destruction of lymphocytes.

Edward [314] studied the cytologic basis of Gordon's encephalitogenic reaction. The agent could be demonstrated in mixtures of myelocytes and older cells from patients with myelocytic leukemia but not in suspensions composed chiefly of myeloblasts. Blood from a patient with eosinophilia gave a strongly positive result. Lymphocytes gave a weakly positive result. Turner, Jackson and Parker [315] demonstrated that the reaction was definitely caused by eosinophils, and this observation was confirmed by McNaught,[316] who supported the theory that Gordon's agent and Friedmann's agent are identical and are apparently derived from the eosinophils and not anything specific for Hodgkin's disease.

---

310. Bonnet, H.; Thieffry, S., and Montefiore: Présence d'un bacille tuberculeux de type aviaire dans un ganglion de lymphogranulomatose maligne, Compt. rend. Soc. de biol. **128**:583, 1938.

311. Donati, A.: Etiologie de la lympho-granulomatose aiguë de Paltauf-Sternberg, Soc. internaz. di microbiol., Boll. d. sez. ital. **9**:318, 1937.

312. Vasconcellos, I.: Encore à propos de l'étiologie de la granulomatose aiguë Paltauf-Sternberg, Soc. internaz. di microbiol., Boll. d. sez. ital. **9**:321, 1937.

313. Decker, F. H.; Leddy, E. T., and Desjardins, A. U.: Leukopenia and Leukocytosis in Lymphoblastoma, Am. J. Roentgenol. **39**:747, 1938.

314. Edward, D. G.: Observations on the Cellular Basis of the Gordon Test for Lymphadenoma, Lancet **1**:936, 1938.

315. Turner, J. C.; Jackson, H., Jr., and Parker, F., Jr.: The Etiological Relation of the Eosinophil to the Gordon Phenomenon in Hodgkin's Disease, Am. J. M. Sc. **195**:27, 1938.

316. McNaught, J. B.: The Gordon Test for Hodgkin's Disease: A Reaction to Eosinophils, J. A. M. A. **111**:1280 (Oct. 1) 1938.

Sachs and Steffel [317] regard the Gordon test as of diagnostic value in Hodgkin's disease but noted that one gland may give a positive reaction whereas another gland from the same patient, with incomplete histologic transformations, may show a negative reaction. In a series of 11 cases of Hodgkin's disease studied by Turner, Jackson and Parker,[315] the Gordon test gave a positive reaction only when eosinophils were present in the glands used in inoculating the rabbits or guinea pigs. The pathogenic agent described by Friedmann and Elkeles in myeloid tissues was shown to be present only in those suspensions of leukocytic cream that contained more than 2,000 eosinophils per cubic millimeter.

Madding [318] analyzed 6 cases of Hodgkin's disease in which there was involvement of the stomach. All the patients were men, 38 to 62 years of age. The red blood cell counts varied from 3,740,000 to 4,000,000 per cubic millimeter. Achlorhydria was present in 1 and fever in none. Clinically the condition simulated that of ulcer or carcinoma of the stomach.

In 5 of the cases, treatment consisted of partial or total excision of the stomach and in 1 case of excision of an ulcer. Three patients were living six to eight years ofter the operation, and 1 was living and well after one year.

Kato and Cardozo [319] studied a 14 year old Negro boy who died of Hodgkin's disease. The red blood cells showed the sickling phenomenon. As the disease progressed, eosinophilia became marked, terminating with 47 per cent eosinophils (26,000 leukocytes). The authors commented on the relation of multinucleated giant cells and megakaryocytes. They favor the concept of myeloid transformation of the lymph nodes, likening the disease in some respects to myelogenous leukemia. Bingold,[320] who considers Hodgkin's disease as an infectious granuloma, described cases in which the tonsils were involved. He suggested that excised tonsils should be studied carefully for incipient Hodgkin's disease, so that wide dissemination can be forestalled by means of radical surgical intervention and energetic roentgen therapy. Falconer and Leonard [321] correlated the incidence and extent of pulmonary involvement with the characteristics and duration of lymphosarcoma, leukemia and Hodgkin's disease. Pulmonary involvement in lymphatic leukemia was 30 per cent, in lymphosarcoma 36 per cent and in Hodgkin's disease

317. Sachs, H. W., and Steffel, W.: Ueber de Bedeutung des Gordon-Testes für die Diagnose der Lymphogranulomatose, Klin. Wchnschr. **17**:1043, 1938.

318. Madding, G. F.: Hodgkin's Disease of the Stomach: Report of Six Cases, Proc. Staff Meet., Mayo Clin. **13**:618, 1938.

319. Kato, K., and Cardozo, W. W.: Hodgkin's Disease with Terminal Eosinophilia Occurring in a Negro Child with Sicklemia, J. Pediat. **12**:165, 1938.

320. Bingold, K.: Die Tonsillen als Eintrittspforte und Sitz der Erkrankung bei Lymphogranulomatose, Deutsches Arch. f. klin. Med. **182**:338, 1938.

321. Falconer, E. H., and Leonard, M. E.: Pulmonary Involvement in Lymphosarcoma and Lymphatic Leukemia, Am. J. M. Sc. **195**:294, 1938.

31 per cent. The duration of life after the onset of pulmonary involvement in these three diseases was two months to eleven years; four months to four and one-half years, and twenty-two to fifty-one months, respectively. Involvement of the pulmonary parenchyma, with pleural effusion, indicated a poor prognosis.

Wright [322] reviewed the symptoms in 60 cases of Hodgkin's disease in which there were intrathoracic lesions. The diagnosis is made by roentgenographic study. The lesions included enlargement of the mediastinal glands (17 patients), involvement of the pulmonary parenchyma (21 patients) and a combination of these. Pleural effusion was present in 17 cases. The average duration of life of 45 patients who died was forty months (seven weeks to twenty-six years). The average length of life of 23 patients who lived longer than this was sixty months, and that of 21 patients who lived less than forty months was twenty months. The average period of survival after the onset of intrathoracic complications was twenty-three months. One patient was apparently cured by surgical removal of enlarged cervical glands (no recurrence in eleven years). For treatment, high voltage roentgen therapy, blood transfusions, mercurial diuretics and improved hygiene and diet have some value. Kasabach and McAlpin [323] found mediastinal involvement in 77 of 251 patients with Hodgkin's disease. Nineteen showed mediastinal involvement roentgenographically without associated peripheral lymphadenopathy. The latter feature became evident on an average of eighteen months later. The average period of survival of the group receiving adequate irradiation was five years and four months. With inadequate treatment the period of survival was three and one-fourth years and with no treatment three and one-twelfth years. A bad prognosis was indicated by generalized distribution of the lesions, osseous invasion, pulmonary and pleural complications and hepatomegaly. In the treatment, maintenance of the patient's weight and general well-being are important. Blasi [324] also commented on the intrathoracic localization of Hodgkin's disease. Varadi's [325] patient had lymphogranulomatous infiltration of the sternum. The diagnosis was made by sternal puncture. In Fonts's [326] patient the lesions of Hodgkin's disease appeared

322. Wright, C. B.: Hodgkin's Disease: Sixty Cases in Which There Were Intrathoracic Lesions, J. A. M. A. **111**:1286 (Oct. 1) 1938.

323. Kasabach, H. H., and McAlpin, K. R.: Mediastinal Hodgkin's Disease, New York State J. Med. **38**:171, 1938.

324. Blasi, R.: Localizzazioni endotoraciche della linfogranulomatose, Quaderni radiol. **2**:123, 1937.

325. Varadi, S.: L'infiltration lympho-granulomateuse du sternum: Sur un case de maladie de Hodgkin, Sang **12**:106, 1938.

326. Fonts Abreau, E.: Un caso de Hodgkin de punto de partida perióstico, Cir. ortop. y traumatol., Habana **5**:277, 1937.

after injury of the periosteum of the sternum. Pessin and Pohle [327] noted an osteomyelitic process of the manubrium and a cutaneous ulcer as the first manifestation of Hodgkin's disease in a 15 year old girl.

The character of the fever in Hodgkin's disease was discussed by Michon and Loth [328] and Michon.[329] In the case reported by Delmas-Marsalet, Bergouignan and Verger [330] the symptoms were caused by compression of the spinal cord by an intraspinal malignant lymphogranuloma. The lesion was not helped by roentgen therapy. In Stewart's [331] case there was marked involvement of the spinal cord. Froment, Croizat and Masson [332] described radiculomedullary compression from a Hodgkin's growth. Symptoms due to marked involvement of glandular groups stimulated several case reports: cervical, Efskind;[333] axillary, Merklen, Gounelle and Warter;[334] rectal, Gallart-Mones and Roca de Vinals.[335] Sherman[336] has summarized the gastrointestinal manifestations in Hodgkin's disease. Osseous lesions marked the cases reported by Herskovits,[337] Abrams [338] and Teenstra.[339] Jutras [340] treated 46 patients (25 males and 21 females) who had pharyngeal lymphosarcoma. Sixteen of these patients lived five years after roentgen

327. Pessin, S. B., and Pohle, E. A.: Hodgkin's Disease with Ulcerative Involvement of the Skin, Am. J. Cancer **34**:220, 1938.

328. Michon, P., and Loth: De la fièvre dite ondulante au cours de la maladie de Hodgkin, Rev. méd. de Nancy **66**:430, 1938.

329. Michon, P.: Fièvre periodique rythmée au cours de la maladie de Hodgkin, Paris méd. **1**:506, 1938.

330. Delmas-Marsalet, Bergouignan and Verger: Compression de la moele par granulomatose intra-rachidienne: Echec de la radiothérapie, Bull. et mém. Soc. méd. et chir. de Bordeaux (1936), 1937, p. 467.

331. Stewart, H. H.: Hodgkin's Disease with Spinal Cord Involvement, Ulster M. J. **7**:68, 1938.

332. Froment, J.; Croizat, P., and Masson, R.: Des compressions radiculo-médullaires dans la granulomatose maligne, J. de méd. de Lyon **19**:71, 1938.

333. Efskind, L.: Ein Beitrag zur Pathologie der Halslymphdrüsen: Unter besonderer Berücksichtigung der "atypischen" Lymphogranulomatose, der Reticuloendotheliosen und der Reticuloendotheliosarkome, Acta path. et microbiol. Scandinav. **15**:16, 1938.

334. Merklen, P.; Gounelle, H., and Warter, J.: Maladie de Hodgkin avec ramollissement des ganglions axillaires, Bull. et mém. Soc. méd. d. hôp. de Paris **53**:1243, 1937.

335. Gallart-Mones, F., and Roca de Vinals, R.: Lymphogranulome de Hodgkin rectal de forme tumorale, Arch. d. mal. de l'app. digestif **28**:67, 1938.

336. Sherman, E. D.: Gastro-Intestinal Manifestations of Lymphogranulomatosis (Hodgkin's Disease), Arch. Int. Med. **61**:60 (Jan.) 1938.

337. Herskovits, E.: Kasuistische Beiträge, Röntgenpraxis **10**:114, 1938.

338. Abrams, H. S.: Osseous System in Hodgkin's Disease, Ann. Surg. **108**: 296, 1938.

339. Teenstra, C. P. H.: Hodgkin's Malignant Granuloma with Marked Skeletal Changes, Nederl. tijdschr. v. geneesk. **82**:391, 1938.

340. Jutras, A.: Protracted Roentgen Therapy of Pharyngeal Lymphosarcoma, Am. J. Roentgenol. **39**:792, 1938.

therapy. The protracted method of Regaud and Coutard was used, with hourly weak dosage and daily exposure (four to twenty-four sittings in sixty-two days). Finzi [341] advocated repetition of submaximal doses of roentgen rays in the treatment of lymphadenoma (Hodgkin's disease). The treatments are first given within two weeks, then repeated in six to eight weeks and then repeated in three months. Repeated submaximal doses are also given to distant parts. "When the disease is localized," he concluded, "the patients are curable, and when it is disseminated, they can be relieved." Craver [342] found that "fewer but larger doses" of roentgen rays are preferable to protracted cycles of fractional doses. Irradiation of the entire body is recommended as an adjunct to local irradiation. Of 121 patients, 17 per cent survived five years or longer. In 220 cases certain symptoms were noted clinically, as follows: pulmonary lesions, 29 per cent; pleural effusion, 17.0 per cent; ascites, 8.0 per cent; jaundice, 6 per cent; itching, 29.0 per cent; cutaneous lesions, 13.8 per cent; herpes zoster, 4.0 per cent; neurologic lesions, 12.0 per cent, and osseous lesions, 18.0 per cent. Sacks [343] presented a case of reticuloendotheliosis and from an analysis of the literature on the subject suggested that the condition is a clinical and pathologic disease entity. It resembles Hodgkin's disease and the aleukemic variety of monocytic leukemia, but morphologic and histologic differences are demonstrable.

LEUKEMIA

Leavell [344] analyzed the records of 136 cases of leukemia in Boston, New York and Philadelphia. For 87 patients with chronic myelogenous leukemia the average duration of life was three and one-fifth years and for 49 patients with chronic lymphatic leukemia three and three-fifths years. Patients with chronic myelogenous leukemia who had a low leukocyte count, marked anemia or evidence of bleeding had a shorter course than those without these symptoms. Marked anemia in lymphatic leukemia indicated an early fatal outcome.

Leukemia, at the extremes of life, has been noted by several authors. Varela and Gambirassi [345] described a subacute blast stage of leukemia

341. Finzi, N. S.: The Roentgen Treatment of Lymphadenoma, Am. J. Roentgenol. **39**:261, 1938.

342. Craver, L. F.: Local and General Irradiation in Hodgkin's Disease. Radiology **31**:42, 1938.

343. Sacks, M. S.: Systemic Proliferation of the Reticuloendothelial System (Reticuloendotheliosis): Report of a Case and Comments on the Literature, Arch. Path. **26**:676 (Sept.) 1938.

344. Leavell, B. S.: Chronic Leukemia: A Study of the Incidence and Factors Influencing the Duration of Life, Am. J. M. Sc. **196**:329, 1938.

345. Varela, M. E., and Gambirassi, A. C.: Mielosis leucémica subaguda a paramieloblastos con metaplasia mieloidea del sistema linfático, en un niño de 18 meses, Arch. argent. de pediat. **8**:1139, 1937.

(myelogenous) in an 18 month old infant. Hart [346] reported data on acute leukemia in childhood, and Schmid [347] and De and Tribedi [348] made similar observations. Simonson [349] analyzed 16 cases of leukemia in childhood. Weil, Isch-Wall, Perlès and Aschkenasy [350] described cases of leukemia in old age.

Spigel [351] analyzed 19 cases of leukemia in children (12, acute lymphatic; 2, chronic lymphatic; 5, acute myelogenous). The ages ranged from 1½ to 14 years, and 60 per cent of the patients were females and 40 per cent males. Spigel considers that leukemia is a condition secondary to a frequently hidden specific infection in hereditarily susceptible persons.

Rhamy [352] studied the blood and tissues of a newborn infant who showed a blood picture with marked erythroblastosis, similar to that in myelogenous leukemia. Death was due to rupture of the enlarged spleen (125 Gm.) during delivery.

From the point of view of causation, Naponen [353] noted points of similarity between myelogenous leukemia and panmyelophthisis. Similar etiologic factors, including severe septic processes and bone marrow poisons (benzene, roentgen rays and radium), are postulated.

Struthers [354] reported the details of 7 cases of acute leukemia in China. Of these, 4 were cases of lymphatic and 3 of myelogenous leukemia. Although leukemia was considered rare in China, the incidence was 1 per six hundred admissions to the medical wards. Evans' [355] summary of the evidence of the neoplastic nature of leukemia was as follows: 1. Cases are cited in which a tumor became evident before changes in the blood occurred. 2. Blood cells in mitosis are seen more frequently in the organs than in the peripheral blood, 3. Invasion of the

346. Hart, F. D.: Acute Lymphatic Leukaemia in Childhood, Lancet **1**:1441, 1938.

347. Schmid, E.: Beitrag zur Klinik und Hämatologie der akuten Leukämien im Kindesalter, Jahrb. f. Kinderh. **151**:149, 1938.

348. De, N. N., and Tribedi, B. P.: Lymphatic Leukemia in Children, Indian J. Pediat. **5**:129, 1938.

349. Simonson, L. M.: Acute Leukemia in Childhood: Analysis of Sixteen Cases, Wisconsin M. J. **37**:110, 1938.

350. Weil, P. E.; Isch-Wall, P.; Perlès, S., and Aschkenasy, A.: La leucémie lymphatique chez les vieillards, Sang **12**:659, 1938.

351. Spigel, H. A.: Leukemias of Infancy and Early Childhood, Arch. Pediat. **55**:7, 1938.

352. Rhamy, B. W.: Leukemia in the New Born, with Death at Birth from Traumatic Rupture of the Spleen, Am. J. Clin. Path. **8**:567, 1938.

353. Naponen, P.: Ueber die Beziehungen der Panmyelophthise zu der akuten myeloischen Leukämie, Acta med. Scandinav., 1938, supp. 89, p. 173.

354. Struthers, E. B.: Acute Leukemia: A Report of Seven Cases, Chinese M. J. **51**:471, 1937.

355. Evans, T. S.: Leukemia: Some Evidence that Leukemia May Be Allied to New Growth, J. Connecticut M. Soc. **2**:112, 1938.

tissues by the leukemic cells occurs. 4. The cells are sensitive to roentgen and radium irradiation. 5. The tendency to skin and bone metastases is marked.

Articles on the relation of industrial poisons, mostly benzene, to the production of leukemia were summarized in the section on "Queries and Minor Notes" in *The Journal of the American Medical Association.*[356] While the disease has appeared occasionally in persons exposed to benzone or radioactive substances, no relation to exposure to war gas has been reported. A case of leukemia following exposure to benzene was described by Perrin, Kissel and Pierquin.[357] A case of benzene poisoning in a patient with malarial splenomegaly (Richon, Vérain, Girard and Devin [358]) and the case of a workman handling betanaphthol (naphthol) (Rolland [359]) were reported. Cantoni [360] described a case of acute aleukemic lymphadenosis following paratyphoid. Lénski [361] reported myelogenous leukemia following splenectomy in a patient with malarial splenomegaly associated with polycythemia. Conen's [362] patient first showed lymphatic leukemia after an infection with Streptococcus viridans.

Maingot, Girard and Bousser [363] studied the blood of a woman who had been a radiologist's assistant for fifteen years and in whom chronic myelogenous leukemia developed. In the third year mild leukocytosis (11,000 to 14,000 leukocytes) was noted. In the eighth year the leukocyte count reached 19,000 and in the eleventh year 95,000; by the thirteenth year it had reached 120,000 per cubic millimeter. Splenomegaly was present. A good result was obtained with roentgen therapy.

Mallick, Ali and Singh [364] noted myeloid leukemia in patients with earlier tuberculous infection. In the case described, marked benefit

356. Industrial Poisons, War Gas and Leukemia, Queries and Minor Notes, J. A. M. A. **110**:1508 (April 30) 1938.

357. Perrin, M.; Kissel, P., and Pierquin, L.: Leucose aiguë benzolique, Paris méd. **1**:533, 1938.

358. Richon, J.; Vérain, M.; Girard, J., and Devin: Anémie splénique avec leucoblastose d'origine benzolique chez un paludéen, Rev. méd. de Nancy **66**:349, 1938.

359. Rolland, R.: Leucémie aiguë chez un ouvrier manipulant du naphtol, Union méd. du Canada **67**:262, 1938.

360. Cantoni, O.: Discussione diagnostica di gravi e oscure emopatie: Linfadenosi acuta leucemica postinfettiva, Haematologica **18**:1019, 1937.

361. Lénski, M.: Myeloid Leukemia Following Splenectomy in a Patient with Malarial Splenomegaly Associated with Polyglobulia, Polska gaz. lek. **17**: 339, 1938.

362. Conen, M.: Zur Streptokokkengenese der lymphatischen Leukämie, Arch. f. Kinderh. **112**:150, 1937.

363. Maingot, G.; Girard, L., and Bousser, J.: Poussées leucocytaires transitoires suivier de leucocytose durable et de leucémie myélogène chez un radiologiste: Contribution à la pathogenie et à la prophylaxie de la leucémie myélogène des radiologistes, Sang **12**:569, 1938.

364. Mallick, S. M. K.; Ali, S., and Singh, B.: Myeloid Leucaemia Treated with Deep X-Ray Therapy, Indian M. Gaz. **73**:19, 1938.

followed high voltage roentgen therapy over the bones. The authors believe that roentgen rays affect the hemopoietic tissue directly and that the neoplastic formation is checked.

Rosenheim [365] described the case of a 56 year old woman in whom pernicious anemia developed. The blood, achlorhydria and symptoms were characteristic, and the response to liver therapy was adequate. The white blood cell count then increased, and typical chronic myelogenous leukemia developed (leukocytes, 93,500 per cubic millimeter). The patient also had tertiary syphilis, but the author considers that the progress of the disease was independent of this and more of the nature of that typical of the disease denoted by the older term leukanemia.

The familial aspects of leukemia have been emphasized by Gottlebe,[366] Boggian [367] and Shipton.[368]

Complications were illustrated by the cases described by Askanazy [369] (diabetes due to leukemic metastases) and by Rachmilewitz [370] (polycythemia).

Teng and Chung [371] found that Leishman-Donovan bodies are phagocytized *in vitro* by neutrophils beyond the myeloblastic stage in leukemic blood and occasionally by eosinophils. Davidsohn [283] noted low titers of isoagglutinins in chronic leukemia, particularly in those cases in which roentgen therapy had not been given. Most patients with acute leukemia or those with chronic leukemia who have received intensive roentgen therapy show normal or slightly elevated titers.

De Lucia and Russo [372] found that in acute leukemia there was a diminution of the total, acid-soluble and lipoid phosphorus fractions, with no change in the inorganic phosphorus. In chronic leukemia all the phosphorus fractions were definitely increased, in spite of the anemia.

---

365. Rosenheim, M. L.: Myeloid Leukaemia Following Pernicious Anemia and Complicated with Tertiary Syphilis, Lancet **2**:1054, 1938.

366. Gottlebe, P.: Ueber familiäres Vorkommen von Leukämie, München. med. Wchnschr. **85**:140, 1938.

367. Boggian, B.: Considerazioni sopra due casi di leucemia familiare, Policlinico (sez. prat.) **45**:472, 1938.

368. Shipton, E. A.: Familial Leucosis, M. J. Australia **1**:116, 1938.

369. Askanazy, M.: Metastasen, die eine neue essentielle Krankheit schaffen (Morbus Addison und Mycosis fungoides, Diabetes und Leukämie), Schweiz. med. Wchnschr. **68**:806, 1938.

370. Rachmilewitz, M.: Polycythaemia and Leukaemia, Harefuah **14**:16, 1938.

371. Teng, C. T., and Chung, H. L.: Phagocytosis of Leishman-Donovan Bodies by Leukemic Blood Cells, Proc. Soc. Exper. Biol. & Med. **39**:156, 1938.

372. de Lucia, P., and Russo, P.: Il quadro del fosforo ematico nelle leucemie, Boll. Soc. ital. di biol. sper. **13**:703, 1938.

Poole and his associates [373] described in detail the history of a man with chronic myelogenous leukemia, covering a period of nine years. He was treated at intervals with solution of potassium arsenite U. S. P., roentgen rays or benzene. The authors commented on the fact that it was easier to maintain the white blood cell count within reasonable limits during the hot summer weather than during the winter, but ultraviolet radiation from artificial sources appeared to do harm rather than good. Chemically, the serum minerals were found to be normal. The values for plasma lipids were increased above normal (lipemia). The plasma phospholipid and free cholesterol components were within the normal range. The neutral fat fraction was high, and the cholesterol esters were strikingly low, suggesting a derangement in fat metabolism. The lipid content of the red blood cells was normal, but the sodium and chloride levels were elevated.

Iwatsuru and Nanjo [374] found a phosphatase content of leukemic blood that was higher than normal, in proportion to the increase in the number of leukocytes. In myelogenous leukemia, phosphatase is liberated into the serum on destruction of the leukocytes, especially after roentgen therapy. The phosphatase content of the urine increases after roentgen therapy.

In lymphatic leukemia Turner, McAlpin and DeLamater [375] reported an increase in the iodine content of the blood, in contrast to the decrease in myelogenous leukemia. Meyer-Bornsen [376] noted that there was not a considerable difference in the sulfur content of the leukocytes in the various types of leukemia. Studies of the bone marrow in leukemia were reported by Tuohy.[377]

Meyer [378] found achlorhydria in 13 per cent of the patients with chronic myelogenous leukemia, in 53 per cent of those with chronic lymphatic leukemia and in 33 per cent of those with aleukemic leukemia. In acute leukemia 2 of 5 had achlorhydria. The average ages of the patients with the different types of leukemia were 44, 60 and 51 and one-half

373. Poole, M. W.; Erickson, B. N.; Williams, H. H.; Burkholder, H. J., and Leucutia, T.: Ten Years of Treatment and Progress in a Case of Chronic Myeloid Leukemia: The Lipid Distribution in Leukocytes and Erythrocytes, J. Michigan M. Soc. **37**:993, 1938.

374. Iwatsuru, R., and Nanjo, K.: Ueber den Gehalt des Blutes an Phosphatase bei der myeloischen Leukämie, Jap. J. M. Sc., VIII, Int. Med., Pediat. & Psychiat. **4**:334, 1938.

375. Turner, K. B.; McAlpin, K. R., and DeLamater, A.: Blood Iodine in Leukemia, Proc. Soc. Exper. Biol. & Med. **39**:55, 1938.

376. Meyer-Bornsen, A.: Ricerche sul solfo organico dei dangue: Il solfo totale dei leucociti nelle leucemie, Biochim. e terap. sper. **24**:479, 1937.

377. Tuohy, E. L.: Bone Marrow Studies, Journal-Lancet **58**:74, 1938.

378. Meyer, O. O.: Achlorhydria in Leucemia, J. Lab. & Clin. Med. **24**:135, 1938.

years, respectively. Isaacs [379] correlated histologic changes in the lymph nodes in various types of lymphopathy with the clinical features corresponding to them.

The neurologic features of leukemia were emphasized in a case described by Nordenson.[380] A man aged 66 years had diabetes and acute aleukemic myelogenous leukemia. Paralysis of the legs developed, followed in two days by paralysis of the hands. The terminal picture was marked by severe anemia, leukocytosis (83,000 cells), a predominance of myeloblasts in the blood and bone marrow, fever, stomatitis and hemorrhagic softening of the central ganglions and the anterior horns of the spinal cord. Gandellini [381] described a case of acute leukopenic leukemia in which neurologic symptoms were prominent. Touw, Nieuwenhuis and Nauta [382] described 2 cases of leukemia characterized by tumor formation. They favor the neoplastic causation of the disease.

Gibson [383] studied the retinal changes in 22 cases of leukemia. The most characteristic features were venous changes, with hemorrhages and exudates. The degree of retinal hemorrhages had a prognostic import. There was close correlation between the amount of retinal hemorrhage and the degree of anemia. Gibson feels that therapy should be directed primarily against the anemia rather than the leukocytosis.

In Aylesworth's [384] case of lymphatic leukemia, unusual ocular symptoms were noted. Cases of chloroma were studied by Frost,[385] Barsoum,[386] Horsfall [387] and Dustin and Thomas.[388] Cookson and MacRae [389] reported a case marked by a lymphoid tumor of the lacrimal

379. Isaacs, R.: Correlation of Clinical and Laboratory Data in Diseases of Lymph Nodes, J. Michigan M. Soc. **37**:1072, 1938.

380. Nordenson, N. G.: Les complications nerveuses des leucémies aiguës, Sang **12**:605, 1938.

381. Gandellini, A.: Contributo allo studio delle sindromi neuroleucemiche: Sudi un caso di leucemia linfatica acuta leucopenica ad insorgenza neurologica, Gazz. d. osp. **59**:650, 1938.

382. Touw, J. F.; Nieuwenhuis, G., and Nauta, J. H.: Two Cases of Leukemia with Tumour-Formation, Acta med. Scandinav. **97**:376, 1938.

383. Gibson, G. G.: Clinical Significance of the Retinal Changes in Leukemia, Arch. Ophth. **20**:364 (Sept.) 1938.

384. Aylesworth, F. A.: Unusual Affection of the Eye in Leukaemia, Canad. M. A. J. **38**:477, 1938.

385. Frost, A. D.: Report of Case with Hematologic Study, Tr. Am. Acad. Ophth. **42**:123, 1937.

386. Barsoum, H.: Case of Chloroma, Brit. M. J. **1**:282, 1938.

387. Horsfall, R. E.: Chloroma: Case History and Postmortem Findings, Brit. M. J. **1**:280, 1938.

388. Dustin, P., Jr., and Thomas, J.: A propos d'un cas de chlorome: Étude anatomo-pathologique et biochimique, Sang **12**:1, 1938.

389. Cookson, H. A., and MacRae, A.: Lymphoid Tumor of the Lacrymal Gland, Brit. J. Ophth. **22**:385, 1938.

gland. Alvis [390] described leukemic infiltration of the retina and choroid in an infant treated with roentgen rays. Dacryocystitis was the first symptom in Stokes' [391] case of lymphatic leukemia in a 68 year old man.

Thiodet's [392] patient with acute lymphatic leukemia (leukocytes, 193,600 per cubic millimeter; blasts, 81 per cent) showed predominately abdominal symptoms. Subocclusion of the intestine appeared to be the cause, possibly as a result of lymphatic hypertrophy or of primary pressure from enlarged mesenteric nodes or secondarily through interference with nerve function. Steinbrinck's [393] patient showed infiltration of the gastric wall. Waitz and Weber [394] reported a case in which phlebitis of the veins of the leg was the first symptom.

Brunner and Schnierer [395] and Brunner and Fischer [396] emphasized the part played by the tonsils in myelogenous and lymphatic leukemia.

Cutaneous manifestations have been described by Goldsmith [397] and by Riehl [398] in myelogenous leukemia and by Kusunoki and Kuwabara,[399] Milbradt [400] and Barney [401] in lymphatic leukemia.

Kost and Rachman [402] described 2 cases of lymphatic leukemia; in 1 case the prominent features were splenomegaly and mediastinal lymphadenopathy, while enlargement of the mesenteric nodes characterized the other. Histologically, however, the features were those of sarcoma.

---

390. Alvis, B. Y.: Leukemic Infiltration of Retina and Choroid in an Infant Treated by X-Ray, Am. J. Ophth. **21**:31, 1938.

391. Stokes, W. H.: Dacryocystitis in Lymphatic Leukemia, Arch. Ophth. **20**: 85 (July) 1938.

392. Thiodet, J.: Leucémie aiguë lymphatique avec syndrome de subocclusion intestinale, Sang **12**:350, 1938.

393. Steinbrinck, W.: Leukämische Infiltration der Magenwand bei lymphatischer Leukämie, Folia haemat. **59**:351, 1938.

394. Waitz, R., and Weber, A.: Leucémie lymphoïde subaiguë ayant débuté par une phlébite du membre inférieur, Strasbourg méd. **98**:231, 1938.

395. Brunner, H., and Schnierer, J.: Die zytologische Untersuchung der Gaumentonsillen und ihre klinische Bedeutung bei Leukämie, Wien. klin. Wchnschr. **50**:1772, 1937; Die Untersuchung der Gaumentonsillen bei Leukämien, Pract. oto-rhino-laryng. **1**:36, 1938.

396. Brunner, H., and Fischer, J.: Die Veränderungen der Gaumentonsillern bei Leukämien, Monatschr. f. Ohrenh. **72**:137, 1938.

397. Goldsmith, W. N.: Chronic Leukaemic Dermatosis, Proc. Roy. Soc. Med. **31**:79, 1937.

398. Riehl, G., Jr.: Chronische leukämische Myelose mit eigenartigen spezifischen Hauterscheinungen, Arch. f. Dermat. u. Syph. **177**:222, 1938.

399. Kusunoki, T., and Kuwabara, S.: Ueber einen Fall von Sclerodermia diffusa, mit Leukämie kompliziert, und durch Parathyreoidektomie gebessert, Arch. f. Dermat. u. Syph. **176**:256, 1937.

400. Milbradt, W.: Klinische Beobachtungen über seltene Hauterscheinungen bei der lymphatischen Leukämie, Dermat. Wchnschr. **105**:1586, 1937.

401. Barney, R. E.: Zosteriform Leukemia Cutis, Arch. Dermat. & Syph. **37**:238 (Feb.) 1938.

402. Kost, E. A., and Rachman, V. I.: Sur la clinique des leucémies atypiques, dites états leucémoïdes, Sang **12**:614, 1938.

In these cases there were possible conditions representing forms of lymphosarcoma cell leukemia.

Aleukemic Leukemia.—Renewed attention has been given to the aleukemic forms of leukemia. Cases have been described by Lindeboom,[403] Fujimoto,[404] Halbron, Lenormand and Jaïs,[405] Bazán and Maggi,[406] Keeler [407] and Christen and Greif.[408] Cabot case 24062 [409] belongs in this group. Miller and Seymour [410] studied 5 cases of leukopenic leukemia and analyzed 18 cases reported in the literature. There was a striking lack of involvement of organs other than the bone marrow. The myeloid tissue showed gross immaturity. Transfusion was the only therapeutic agent of at least temporary value. Weil and Aschkenasy [411] described a case of "cryptoleukemia" with an aleukemic blood picture and no splenomegaly or glandular enlargement. The case appeared to be one of severe aplastic anemia, but sternal puncture showed its true nature—aleukemic lymphatic leukemia.

Monocytic Leukemia.—Montgomery and Watkins [412] described 4 cases of monocytic leukemia, Schilling type, in which exfoliative dermatitis was a manifestation. A case of monocytic leukemia in a man aged 39 years was described by Beck.[413] The peculiar configuration of the rosette of vacuoles in the *Hof* of the nucleus, on supravital staining, was

403. Lindeboom, G. A.: Ueber die sogenannte aleukämische megakaryocytäre Myelose, Acta med. Scandinav. **95**:388, 1938; So-Called Aleukemic Megakaryocystic Myelosis, Nederl. tijdschr. v. geneesk. **82**:3072, 1938.

404. Fujimoto, M.: Ein Fall von acuter myeloischer Leukämie mit geringer Leukozytenzahl im ganzen Verlauf, Nagasaki Igakkai Zassi **16**:1006, 1938.

405. Halbron, P.; Lenormand, and Jaïs: Un nouveau cas de leucose aiguë ayant pris le masque d'une aleucïe hémorragique: Les difficultés d'interprétation du myélogramme, Bull. et mém. Soc. méd. d. hôp. de Paris **54**:56, 1938.

406. Bazán, F., and Maggi, R.: Sobre un caso de linfadenosis aguda aleucémica, Arch. argent. de pediat. **9**:385, 1938

407. Keeler, H. R.: Acute Hypocytic Lymphatic Leukemia, Internat. Clin. **3**:271, 1938.

408. Christen, W., and Greif, S.: Aleukämische Lymphadenose mit hyperchromer megalozytärer Anämie: Vergleichende Untersuchungen von Sternalmark, Melleolarmark, und peripherem Blut, Wien. Arch. f. inn. Med. **32**:85, 1938.

409. Aleukemic Lymphatic Leukemia, Cabot Case 24062, New England J. Med. **218**:270, 1938.

410. Miller, F. R., and Seymour, W. B.: Leukopenic Leukemia of the Myeloblastic Type, Am. J. M. Sc. **196**:621, 1938.

411. Weil, P. E., and Aschkenasy, A.: Un cas de crypto-leucémie lymphatique sans splénomégalie ni adénopathies simulant une anémie grave aplasique, Sang **12**:359, 1938.

412. Montgomery, H., and Watkins, C. H.: Exfoliative Dermatitis as a Manifestation of Monocytic Leukemia (Schilling), Proc. Staff Meet., Mayo Clin. **13**: 294, 1938; Minnesota Med. **21**:636, 1938.

413. Beck, R. C.: Monocytic Leukemia with Analysis of Cell Characteristics by Supravital and Fixed Staining Technics, Am. J. Clin. Path. **8**:509, 1938.

confirmed as a specific test of the moncytic cells. Plum and Thomsen,[414] in describing 3 cases of monocytic leukemia, noted that this disease may be confused at times with infectious mononucleosis or with agranulocytosis. They emphasized the points in the differential diagnosis. Bouchut, Guichard, Moreau and Mathieu [415] described a case of monocytic leukemia with an acute evolution. In cases of monocytic leukemia Wyckoff [416] reported the results of studies of the cells in the blood or bone marrow by means of supravital stains. The monocytic cells showed a nucleus with reticular chromatin and one or two nucleoli. The peroxidase test gave reactions which varied from heavy to negative. Newns and Signy [417] reported on the clinical and histologic aspect of monocytic leukemia in 2 children, 2 and 2½ years old, respectively. The course was acute, with death in ten and six ond one-half days, respectively. Waitz and Hoerner [418] described an aleukemic leukemoid blood picture in a man whose bone marrow contained many large reticulum cells. The internal organs and the eyes showed hyperplasia of the "reticuloendothelial" cells. Symmers [419] described 4 cases of giant follicular lymphoblastoma. Clinically the symptoms resembled those of Hodgkin's disease, certain forms of lymphosarcoma and lymphatic leukemia. Histologically there was dimensional hyperplasia of the lymph follicles of the lymph nodes and spleen. The author considers the condition to be of "toxic" or "inflammatory" origin and amenable to mild roentgen therapy. Transformation to polymorphous cell sarcoma, Hodgkin's disease and the changes of lymphatic leukemia may be sequelae. The previously undescribed phase of the evolution of the disease is necrotic folliculitis of the lymph nodes and spleen.

Isaacs [420] placed the cell called promyelocyte in the monocytic series and described it as typical of the stage after the monocyte blast (metamonoblast).

---

414. Plum, P., and Thomsen, S.: Three Cases of Monocytic Leukaemia, Acta med. Scandinav. **96**:578, 1938.

415. Bouchut, L.; Guichard, A.; Moreau, and Mathieu: Leucémie à monocytes d'évolution aiguë, Lyon méd. **161**:317, 1938.

416. Wyckoff, H. A.: Monocytic Leukemia: Some Blood and Bone Marrow Studies, California & West. Med. **48**:348, 1938.

417. Newns, G. H., and Signy, A. G.: Two Cases of Monocytic Leukaemia, Proc. Roy. Soc. Med. **31**:364, 1938.

418. Waitz, R., and Hoerner, G.: Syndrome agranulocytaire avec myéloblestémie et prolifération réticulo-endothéliale médullaire, viscéral, oculaire: Intérêt diagnostique de cette prolifération, Sang **12**:801, 1938.

419. Symmers, D.: Giant Follicular Lymphadenopathy With or Without Splenomegaly: Its Transformation into Polymorphous Cell Sarcoma of the Lymph Follicles and Its Association with Hodgkin's Disease, Lymphatic Leukemia and an Apparently Unique Disease of the Lymph Nodes and Spleen—a Disease Entity Believed Heretofore Undescribed, Arch. Path. **26**:603 (Sept.) 1938.

420. Isaacs, R.: Diagnosis of Leukemia and Response to Therapy, Univ. Hosp. Bull., Ann Arbor **4**:129, 1938.

Leukemia and Pregnancy.—Erf and Fine [421] reported the thirty-first case recorded in the literature of a pregnant woman with leukemia. This patient with chronic myelogenous leukemia received intensive roentgen therapy while she was five and one-half to six months pregnant. Six weeks later labor was induced surgically and a normal boy delivered. The mother died seven months later in the stage of myeloblastic leukemia. Although the uterus received at least 200 roentgens, the child did not show any ill effects. Repeated studies of the bone marrow were made on the mother and child.

Forkner [422] discussed the relation of leukemia to pregnancy and the puerperium.

Treatment of Leukemia.—Cooke [423] treated patients with acute leukemia with extracts of fresh red bone marrow. The patients also received blood transfusions and other symptomatic treatment. Two patients had complete remissions for from two to four months; 2 had remissions for shorter periods; 2 showed an increase in the maturation stage of the polymorphonuclear cells; 4 showed arrest of the disease clinically for from three to six months, and 2 showed a decrease in the size of the leukemic lymph nodes and spleen. Other patients, however, showed no effect. The author believes that the course of the disease was influenced definitely in some cases but gave a rather guarded opinion as to the part played by the marrow extract.

For the aleukemic forms of leukemia, Fricke and Watkins [424] recommend radium therapy over the region of the spleen rather than roentgen therapy. Fifteen of 16 patients (leukopenic phases of chronic myeloid and lymphatic leukemia, monocytic leukemia and subleukemic splenic reticuloendotheliosis) showed improvement in the condition of the blood and symptoms. Gosio [425] transfused blood from patients with myelogenous leukemia into patients with lymphatic leukemia and vice versa. When normal blood was used in a transfusion to a patient with lymphatic leukemia, the number of red blood cells increased; when blood from a patient with lymphatic leukemia was given to a patient with lymphatic

421. Erf, L. A., and Fine, A.: Serial Blood and Bone Marrow Findings of an Eight Month Premature and Its Roentgen Ray Treated Chronic, Myeloid Leukemic Mother, Am. J. M. Sc. **195**:8, 1938.

422. Forkner, C. E.: The Relationship of Leukemia to Pregnancy and the Puerperium, Internat. Clin. **2**:29, 1938.

423. Cooke, J. V.: Experimental Therapy of Acute Leukemia with Extracts of Bone Marrow, J. Pediat. **13**:651, 1938.

424. Fricke, R. E., and Watkins, C. H.: The Radium Treatment of Rare Forms of Leukemia, Proc. Staff Meet., Mayo Clin. **13**:193, 1938; Minnesota Med. **21**:96, 1938.

425. Gosio, R.: Le trasfusioni interleucemiche dal punto di vista della regolazione biologica dei tessuti iperleucoplastici, Policlinico (sez. med.) **45**:285, 1938; Ulteriore contributo allo studio della fisiopatologia del leucemico: Ricerche complementari alle transfusioni interleucemiche, ibid. **45**:42, 1938.

leukemia, the leukocytosis became more marked. When the blood from a patient with myelogenous leukemia was used, the leukocytosis remained unchanged. The myeloid cells disintegrated, and their products disappeared from the blood stream in from eighteen to twenty hours. The lymph nodes served as the filtering mechanism. A number of other transfusion experiments with leukemic blood have been described. Galán Conesa and Montero [426] used polycythemic blood in treating leukemia. Ravina [427] used leukemic blood in the treatment of agranulocytosis. A lasting therapeutic result was noted only in their last patient.

Mauro [428] noted little therapeutic effect when ascorbic acid was given to patients with Werlhof's disease, myelogenous leukemia or acute or chronic lymphatic leukemia. The amount of ascorbic acid in the urine gave no definite information about the condition in these diseases. Thiele [429] found no lasting effect on the leukocyte count when vitamin C was given intravenously, although there was a temporary increase in the number. The medication had no effect on chronic myelogenous leukemia.

Leukemoid Conditions.—DuBois [430] described the leukemoid condition produced in a man of 53 years by carcinoma of the bladder with metastasis to the lungs and prostate. The leukocyte count was 119,437 per cubic millimeter, with 73.75 per cent mature polymorphonuclear neutrophils. There were no metastases to the bone marrow.

An eosinophilic leukemoid reaction appeared in Recchia's [431] case of acute colitis. In Leibowitz' [432] patient, tuberculous sepsis was accompanied by a myeloblastic blood picture.

Leukemia in Animals.—Morton and Mider [433] noted lymphomatosis in 10 of 48 mice painted with methylcholanthrene in solution in commercial benzene. In 2 cases the leukocyte count rose to 90,000 and 139,000

426. Galán Conesa, E., and Montero, R.: Contribución al estudio de las trasfusiones de globulos: Un caso de leucemia dinfoide temporalmente convertido en policitémico, Bol. Soc. cubana de pediat. **9**:547, 1937.

427. Ravina, A.: Agranulocytose: Traitement par la transfusion de sang leucémique, Presse méd. **45**:1760, 1937.

428. Mauro, E.: L'acido ascorbico in alcune emopatie, Minerva med. **2**:613, 1938.

429. Thiele, W.: Die Wirkung des Vitamin C auf das weisse Blutbild und die chronische myeloische Leukämie, Klin. Wchnschr. **17**:150, 1938.

430. DuBois, A. H.: Leucocytose carcinomateuse pseudo-leucémique, Sang **12**: 317, 1938.

431. Recchia, F.: Reazione leucemoide ad impronta eosinufila in corso di colite acuta, Policlinico (sez. prat.) **45**:369, 1938.

432. Leibowitz, S.: Tuberculous Sepsis with Myeloblastic Blood Picture, Arch. Path. **25**:365 (March) 1938.

433. Morton, J. J., and Mider, G. B.: The Production of Lymphomatosis in Mice of Known Genetic Constitution, Science **87**:327, 1938.

per cubic millimeter, respectively. No tumors appeared in 50 control mice.

Lewis [434] was able to transplant, through twelve generations of mice, three sarcomas induced by the injection of 1, 2, 5, 6-dibenzanthracene. The tumors could not be propagated by means of the injection of blood plasma or with centrifuged tumor extracts.

Lits, Kirschbaum and Strong [435] noted regression of a malignant lymphoid tumor in mice, with prolongation of life (fifty and one-half instead of thirty-one and one-half days) after the injection of colchicine. A series of "caryoclasic shocks," with pyknosis and death of the lymphocytes of the tumor, was observed. Normal thymic lymphocytes were also destroyed. Engelbreth-Holm and Frederiksen [436] produced leukemia in 36 of 59 mice 4 weeks old by the injection of a cell-free extract of tissues from leukemic mice. The material was prepared by a special anaerobic technic, being reduced with cobalt-cysteine. This experiment seemed to indicate that mouse leukemia, like fowl leukemia, may be transmitted by a chemical rather than a cellular medium. These authors [437] concluded that the agent of fowl leukosis is inactivated by oxidation. If the process is interrupted before it is complete, it is possible to reactivate the material almost to its original potency with a cysteine cobalt sulfate reducing system. These data are interpreted to favor a chemical rather than a living (micro-organism) causative agent of fowl leukosis.

Potter, Taylor and MacDowell [438] were able to transmit immunity to mouse leukemia from cell-immunized mice to normal mice by implantation of tissue from actively immunized mice. Hall and Knocke [439] were able to transmit chloroleukemia in mice by the intravenous injection of a suspension of leukemic cells. While subcutaneous inoculation produced a slowly growing tumor, intravenous injection was followed by generalized leukemia. The greenish color of the lymph nodes was not

434. Lewis, M. R.: Transplantable Lymphosarcoma in Mice, Am. J. Cancer **34**:399, 1938.

435. Lits, F. J.; Kirschbaum, A., and Strong, L. C.: Action of Colchicine on a Transplanted Malignant Lymphoid Neoplasm in Mice of the C3H Strain, Am. J. Cancer **34**:196, 1938; Action of Colchicine on a Malignant Lymphoid Neoplasm in Mice of an Inbred Strain, Proc. Soc. Exper. Biol. & Med. **38**:555, 1938.

436. Engelbreth-Holm, J., and Frederiksen, O.: The Transmission of Mouse-Leucaemia to Healthy Animals by Means of Cell-Free Substance, Acta path. et microbiol. Scandinav., 1938, supp. 37, p. 145.

437. Engelbreth-Holm, J., and Frederiksen, O.: The Reactivation of the Fowl-Leukosis Agent After Inactivation by Oxydization, Acta path. et microbiol. Scandinav., 1938, supp. 37, p. 138.

438. Potter, J. S.; Taylor, M. J., and MacDowell, E. C.: Transfer of Acquired Resistance to Transplantable Leukemia in Mice, Proc. Soc. Exper. Biol. & Med. **37**:655, 1938.

439. Hall, J. W., and Knocke, F. J.: Transmission of Chloroleukemia of Mice, Am. J. Path. **14**:217, 1938.

due to eosinophils. After exposures to roentgen rays, the immunity of mice to subcutaneous inoculations was decreased.

Breedis and Furth [440] were able to preserve mouse leukemic cells at —70 C. for as long as four hundred and thirty days and produce lesions on intravenous or subcutaneous inoculations. The types used included lymphoid leukemia (eight to four hundred and forty days), myelocytic leukemia (twenty to four hundred and forty days), chloroleukemia (one to thirteen days) and monocytic leukemia (two to four hundred and thirty days). Breedis and Furth consider that it is the living leukemic cell which is preserved, and not only a virus. They noted that the cells can be inactivated by roentgen rays while in the frozen state, a feature not characteristic of a virus. Bichel [441] was able to cultivate cells of mouse leukemia in vitro for at least five months by adding actively growing cultures of fibroblasts. A single addition of fibroblasts enabled the cells to grow actively for ten to twelve generations. Division was by mitosis. The cells were easily injured but when injected into mice produced the disease. Yudina [442] gave weekly injections of a 0.5 per cent solution of 1, 2, 5, 6-dibenzanthracene subcutaneously to 45 chickens for five months. Of these, 1 had leukemia, 3 had aleukemic myelosis, 1 of which also had sarcoma, and 1 had uncomplicated sarcoma. The condition in 1 chicken recalled that of a patient of Ahlstrom's [443] who had reticular cell sarcoma and lymphatic leukemia.

Clapham [444] observed that leukemic fowl showed forty times heavier infestation with tapeworm than normal fowl. Clapham interpreted this to indicate a reduced resistance to tapeworm as a manifestation of the leukemia. Rask-Nielsen [445] studied the transmission of a neoplasm which consisted of pathologic myeloblasts. The white rats died after about twenty-three days and showed multiple tumors in the abdominal organs. Stasney and Feldman [446] studied the evolution of leukemic lymphoblastoma in a 3 month old calf. There was general lymphadenopathy, and the histologic details of the tissues resembled those of lymphatic leukemia of man. The leukocyte count was 24,000 per cubic milli-

440. Breedis, C., and Furth, J.: The Feasibility of Preserving Neoplastic Cells in the Frozen State, Science **88**:531, 1938.

441. Bichel, J.: Dauerzüchtung von leukämischen Zellen in vitro, Ztschr. f. Krebsforsch. **48**:92, 1938.

442. Yudina, N. D.: Leukosis of Chickens Induced by Injections of Cancerigenic Agent *1: 2: 5: 6-di*Benzanthracene, Med. zur. **7**:819, 1937.

443. Ahlstrom, C. G.: Gleichzeitiges Vorkommen eines Retikelzellsarkoms und einer lymphatischen Leukämie, Virchows Arch. f. path. Anat. **301**:49, 1938.

444. Clapham, P. A.: The Relation of Helminthiasis to Leukaemia in Domestic Fowls, J. Helminthol. **16**:53, 1938.

445. Rask-Nielsen, R.: Experimental Studies on a Transplantable Aleukemic Myelomatosis in White Rats, Acta path. et microbiol. Scandinav. **15**:285, 1938.

446. Stasney, J., and Feldman, W. H.: Leukemic Lymphoblastoma in a Calf: A Hematologic and Histologic Study, Am. J. Cancer **34**:240, 1938.

meter, with 68 per cent immature lymphocytes. Furth and Furth [447] noted monocytic leukemia in 9 of 96 mice which were given 1, 2 benzpyrene intrasplenically. The incidence of myelogenous leukemia was greater than that in the controls, although a causal relation was not definite. The Rask-Nielsens [448] were unable to demonstrate a specific agent by the injection of killed leukotic cells into irradiated and non-irradiated mice. Lanza [449] produced myelogenous leukemia in rats by the injection of 1, 2 benzpyrene into the bone marrow; Pikovski and Doljanski [450] reported similar results.

The basal metabolic rate was found by Olson and Dukes [451] to be slightly increased in transmissible fowl leukosis and fibrosarcoma and greatly increased in lymphosarcoma.

Victor and Potter [452] found that the serum dextrose value (heart blood) for leukemic mice (ether anesthesia) varied from 38 to 129 mg. per hundred cubic centimeters, as compared with 159 to 284 mg. in normal and immune mice. Neither the administration of dextrose nor that of adrenal cortical extract prolonged the life of leukemic mice. These authors [453] also found that the respiratory rate of normal lymphatic tissue and several transmission lines of lymphatic leukemia is depressed by dextrose. The amount of depression varied with the transmission line. There appeared to be no correlation between the changes in the respiratory quotient produced by dextrose and in the glycolytic rates of cells of lymphatic leukemia tissue.

Victor and Potter [454] noted that when 0.2 per cent dextrose was added to cells of mouse lymphatic leukemia in leukemic serum, anaerobic glycolysis was about three times greater than when normal serum was used. When no dextrose was added, the oxygen consumption was

---

447. Furth, J., and Furth, O. B.: Monocytic Leukemia and Other Neoplastic Diseases Occurring in Mice Following Intrasplenic Injection of 1:2-Benzpyrene, Am. J. Cancer **34**:169, 1938.

448. Rask-Nielsen, H. C., and Rask-Nielsen, R.: Further Studies on a Transmissible Myeloid Leukosis in White Mice, Acta path. et microbiol. Scandinav. **15**:169, 1938.

449. Lanza, G.: Quadri leucemici nei ratti da inienzioni intramidollari di 1:2 benzopirene: Effetti della inoculazione di sangue ed organi di ratti con leucemie da 1:2 benzopirene in ratti normali, Pathologica **30**:185, 1938.

450. Pikovski, M., and Doljanski, L.: Leukaemia Produced by Cancerogenic Substance: Preliminary Note, Harefuah **14**:xi, 1938.

451. Olson, C., and Dukes, H. H.: The Basal Metabolic Rate of Chickens Affected with Fowl Paralysis, Transmissible Fowl Leucosis and Certain Spontaneous Neoplasms, Folia haemat. **60**:57, 1938.

452. Victor, J., and Potter, J. S.: Low Serum Glucose in Leucemic Mice, Am. J. Cancer **33**:578, 1938.

453. Victor, J., and Potter, J. S.: The Respiratory Quotients of Normal and Leukemic Mouse Lymphoid Tissue, Am. J. Cancer **32**:554, 1938.

454. Victor, J., and Potter, J. S.: Leukemic Cell Metabolism in Serum of Normal, Immunized and Leukemic Mice, Am. J. Cancer **33**:568, 1938.

greater than the carbon dioxide elimination. This is just the reverse of the condition in normal or immune serum. In normal serum, leukemic and normal lymphoid cells have higher respiratory rates than in Ringer's solution, while the anaerobic glycolytic rates are the same in the two mediums. Butler and Warren [455] noted quick recovery from paralysis and a return of the blood picture to normal in leukemic fowls given injections of 1 or 2 cc. of cold pressed wheat germ oil in the breast. Cole,[456] however, was unsuccessful in this respect when he injected wheat germ oil intraperitoneally.

NIEMANN-PICK DISEASE, SCHÜLLER-CHRISTIAN DISEASE, GAUCHER DISEASE, AND LIPOID HISTIOCYTOSIS

An extensive review of the clinical and physiologic features of the xanthomatous diseases, including Niemann-Pick disease, Gaucher's disease and Hand-Schüller-Christian disease, was given by Thannhauser and Magendantz.[457] The data are illustrated with material on 22 cases, and a bibliography of one hundred and fifty-nine articles is given. General consideration of the pathologic features of the lipoidoses has also been given by Epstein [458] and Müller.[459]

Naegeli [460] analyzed the geographic distribution of various hemopoietic dyscrasias. In addition to the more common diseases, data on some of the more unusual forms were given. A group of over 100 cases of ovalocyte anemia were reported from Danzig, and 11 cases of Hand-Schüller-Christian disease were reported from Debrecen, Hungary.

Sternal puncture material may be of aid in the diagnosis of Gaucher's disease. Schartum-Hansen [461] demonstrated the characteristic cells in the marrow of 2 patients. The marrow itself was hyperplastic, with a relative increase in "macroblasts" and an increase in the relative percentage of erythroblasts as compared with that of the leukopoietic cells. Erf [462]

455. Butler, W. J., and Warren, D. M.: Fowl Leucemia and Vitamin E: A Preliminary Report, J. Am. Vet. M. A. **45**:204, 1938.

456. Cole, R. K.: Vitamin E and Avian Neurolymphomatosis, Science **88**: 286, 1938.

457. Thannhauser, S. J., and Magendantz, H.: Different Clinical Groups of Xanthomatous Diseases: A Clinical Physiological Study of Twenty-Two Cases, Ann. Int. Med. **11**:1662, 1938.

458. Epstein, E.: Beiträge zur Pathologie der allgemeinen Lipoidosen, Ergebn. d. allg. Path. u. path. Anat. **33**:280, 1937.

459. Müller, H.: Ueber die sogenannten primären Lipoidosen, Ztschr. f. Kinderh. **59**:476, 1938.

460. Naegeli, O.: Geographisch-medizinische Erforschung der Anaemien, in Comptes rendus de la troisieme Conférence international de pathologie géographique, Helsingfors, Mercators Tryckeri, 1937, p. 81.

461. Schartum-Hansen, H.: Sternalpunktion bei Morbus Gaucher, Folia haemat. **61**:180, 1938.

462. Erf, L. A.: Studies of Gaucher Cells by the Supravital Technique, Am. J. M. Sc. **195**:144, 1938.

found Gaucher cells in sternal puncture material from 5 patients; 1 patient had myelocytes and myeloblasts in the blood stream, 1 had no palpable enlargement of the liver or spleen and 1 was severely jaundiced. On supravital staining, the Gaucher fibrils in the cell cytoplasm were often S shaped, with tapered extremities; they were 5 to 10 microns in length and 0.5 to 1.0 microns in width.

Clinical and chemical studies of Gaucher's disease are reported by Zehnder [463] and by Pachman.[464] Webster [465] described a case of Gaucher's disease in a 6 year old girl. The spleen was removed. Six and a half years later the patient appeared well, although there was some increase in pigmentation. The patient reported on by Pack and Silverstone [466] also showed improvement after splenectomy.

Freudenberg [467] made studies of the blood and tissues of identical twins dying of Niemann-Pick disease. Increased esterase and high phosphatase values were found for the blood serum. The findings did not suggest that the lipoid abnormality depended on deficient enzyme action.

Teunissen and den Ouden [468] concluded that the phosphatide stored in the spleen in Niemann-Pick disease is sphingomyelin and that the lecithin increase is secondary and smaller. Cases were described by Lignac and Teunissen,[469] Loebell [470] and Atkinson.[471]

Kato [472] studied a 9 month old girl with Niemann-Pick disease. Symptoms were noted at the age of 3 months. Dried splenic substance showed 3.08 per cent lecithin. From the spleen in Niemann-Pick

463. Zehnder, M.: Klinischer und chemischer Beitrag zum Studium des Morbus Gaucher, Deutsche Ztschr. f. Chir. **250**:422, 1938.

464. Pachman, D. J.: Chronic Gaucher's Disease, Am. J. Dis. Child. **56**:248 (Aug.) 1938.

465. Webster, R.: Pathological Reports from Children's Hospital, Melbourne: V. Gaucher's Disease, M. J. Australia **2**:22, 1938.

466. Pack, G. T., and Silverstone, S. M.: Gaucher's Disease: Report of a Case Improved After Splenectomy, Am. J. Surg. **41**:77, 1938.

467. Freudenberg, E.: Klinische Beobachtungen und Untersuchungen an einem Zwillingspaar mit Niemann-Pickscher Krankheit (NPK), Ztschr. f. Kinderh. **59**:313, 1937.

468. Teunissen, P. H., and den Ouden, A.: Contributions to the Study of Phosphatide Lipoidosis (Niemann-Pick's Disease), Nederl. tijdschr. v. geneesk. **84**:2406, 1938.

469. Lignac, G. O. E., and Teunissen, P. H.: Beitrag zur Kenntnis der Lipoidosis phosphatidica, Beitr. z. path. Anat. u. z. allg. Path. **101**:139, 1938.

470. Loebell, H.: Niemann-Picksche Erkrankung und Ohr, Hals-, Nasen- u. Ohrenarzt (Teil 1) **29**:119, 1938.

471. Atkinson, F. R. B.: Niemann-Pick's Disease, Brit. J. Child. Dis. **34**: 245, 1937.

472. Kato, S.: A Case of Niemann-Pick's Disease, Acta pædiat. japon. **44**: 533, 1938.

disease, Chargaff [473] isolated a substance which had marked anticoagulant properties. Chemically the material was a sulfuric acid ester. Wood [474] described a case of Niemann-Pick disease in an 8 month old white girl. At autopsy there was, in addition to the generalized xanthomatosis, primary carcinoma of the liver (hepatic cell type).

Kennedy [475] summarized the clinical features of 8 cases of Schüller-Christian disease. The xanthomatous changes were widespread in the organs and tissues, and the skull was involved in all the cases. The blood lipoid values did not vary significantly from normal. Rapid healing followed radium or roentgen therapy. Cases of Schüller-Christian disease were described by Aboulker, Bertrand-Guy and Faraggi,[476] Benedek,[477] Hallervorden,[478] Marinescu, Draganesco, Stroesco and Palade,[479] Vampre, Villaca and Delape,[480] Miyaji,[481] Shea,[482] van Bogaert,[483] Warburg,[484] Weissenborn and Wurm,[485] Wynkoop and Hadley [486] and Hankey.[487]

---

473. Chargaff, E.: Studies on the Chemistry of Blood Coagulation: VIII. Isolation of a Lipid Inhibitor of Blood Clotting from the Spleen in a Case of Niemann-Pick's Disease, J. Biol. Chem. **125**:677, 1938.

474. Wood, H.: Generalized Essential Xanthomatosis (Type Niemann-Pick) Associated with Primary Carcinoma of the Liver in an Infant, Arch. Path. **26**: 873 (Oct.) 1938.

475. Kennedy, R. L. J.: Xanthomatosis: Schüller-Christian's Disease, Proc. Staff Meet., Mayo Clin. **12**:776, 1938.

476. Aboulker, H., Bertrand-Guy and Faraggi: Deux cas de maladie de Schüller-Christian, Bull. et mém. Soc. de radiol. méd. de France **25**:721, 1937; Algérie méd. **42**:36, 1938.

477. Benedek, L.: Hand-Schüller-Christian Syndrome, Orvosi hetil. **82**:211, 1938.

478. Hallervorden, J.: Gehirnbefunde bei Christian-Schüllerscher Krankheit und allgemeinen Cholesterinosen, Ztschr. f. d. ges. Neurol. u. Psychiat. **161**:384, 1938.

479. Marinescu, G.; Draganesco, S.; Stroesco, G., and Palade, G.: Examen anatomo-clinique d'un cas atypique de la maladie de Schüller-Christian, Ann. d'anat. path. **14**:673, 1937.

480. Vampre, E.; Villaca, C. M., and Delape, J. B.: Molestia de Hand-Schüller-Christian, Rev. Assoc. paulista de med. **11**:293, 1937.

481. Miyaji, S.: Experimentelle Beiträge zur Frage der xanthomatösen Erkrankungen, Arch. f. klin. Chir. **192**:190, 1938.

482. Shea, J. J.: Xanthomatosis (Schüller-Christian's Disease): Report of a Case with Radiosensitive Pathology in Mastoid, Laryngoscope **48**:589, 1938.

483. van Bogaert, L.: Les aspects neurologiques des cholestérinoses généralisées, Bull. Acad. roy. de méd. de Belgique **3**:206, 1938; Progrès méd., May 28, 1938, p. 785.

484. Warburg, E.: Case of Hand-Schüller-Christian's Disease, Ugesk. f. læger **100**:749, 1938.

485. Weissenborn and Wurm, H.: Lipoidgranulome des Schädeldachs ohne allgemeine Lipoidgranulomatose (Hand-Schüller-Christiansche Krankheit) bei generalisierter Tuberkulose, Chirurg **10**:462, 1938.

(Footnotes continued on next page)

BONE MARROW

Weil and Perlés [488] published an illustrated monograph on the findings for different diseases on sternal puncture. Reiter [489] called attention to the nonhomogeneous distribution of the marrow in the sternum in aplastic anemia, granulocytopenia and myelophthisis, as compared to the uniform changes in leukemia and pernicious anemia. In those cases in which there is irregular distribution of the marrow, sternal puncture may give misleading pictures and result in misinterpretation. Osgood [490] proposed a nomenclature for blood and bone marrow cells. The differential points in the identification of each type of cell were given in analytic tables. The new names differ from the standard nomenclature (e. g., red blood cell is akaryocyte, and polymorphonuclear neutrophil is lobocyte), but all hematologists have not yet accepted the changes.

Cultures of bone marrow cells were used by Osgood [491] to determine the method of action of sulfanilamide. The drug had no effect on phagocytosis but appeared to neutralize the toxins of beta hemolytic streptococci.

Huggins and Smith [492] found that intravenous injections of colloidal thorium dioxide caused no significant changes in the cellular elements of venous blood. Thorium was fixed by the cells of the reticuloendothelial system. Anemia or plethora did not cause migration of the cells, and in newly formed marrow the macrophages were free from thorium. Growth in the thickness of the bone marrow occurs at its circumference.

Fitz-Hugh,[493] in summarizing the action of certain drugs on the blood and bone marrow, noted reports of aplastic anemia (I), hemolytic anemia (II), thrombopenic purpura (III), agranulocytic angina (IV)

---

486. Wynkoop, E. J., and Hadley, L.: Schüller-Christian's Disease: Report of a Case, Arch. Pediat. **55**:417, 1938.

487. Hankey, G. T.: Three Unusual Affections of the Jaw: Fibro-Myxo-Sarcoma of the Mandible; Multilocular Cyst Arising from Maxillary Dental Cyst; Xanthomatosis or Lipoid Granulomatosis of the Mandible (Schüller-Christian's Syndrome), Proc. Roy. Soc. Med. **31**:1137, 1938.

488. Weil, P. E., and Perlés, S.: La ponction sternale: Procédé de diagnostic cytologique, Paris, Masson & Cie, 1938.

489. Reiter, B.: Anatomische Untersuchungen zur Frage der Inhomogenität des Knochenmarkes im Hinblick auf die Auswertung der Sternalpunktion, Ztschr. f. d. ges. exper. Med. **103**:694, 1938.

490. Osgood, E. E.: The Histogenesis, Classification and Identification of the Cells of the Blood and Marrow Based on Cultures and Hematologic Studies of Human Marrow and Blood, Am. J. Clin. Path. **8**:59, 1938.

491. Osgood, E. E.: Culture of Human Marrow: Studies on the Mode of Action of Sulfanilamide, J. A. M. A. **110**:349 (Jan. 29) 1938.

492. Huggins, C., and Smith, K. M.: The Effect of Hypertrophic Cartilage on Bone Marrow Growth, J. Exper. Med. **67**:41, 1938.

493. Fitz-Hugh, T.: Sensitivity Reactions of the Blood and Bone Marrow to Certain Drugs, J. A. M. A. **111**:1643 (Oct. 29) 1938.

and leukemoid reactions (V) after the use of the following substances:[494] acetanilid (II), aminopyrine (II to V), antipyrine (II), arsphenamine (I to V), benzene (I to V), bismuth (III, IV), causalin (IV), cinchophen dinitrophenol (IV), ergot (III), gold salts (I, III, IV), neostibosan (IV), nirvanol (IV), novaldin (IV); acetophenetidin (IV), plasmochin (IV), quinine (II to V), sedormid (III) and sulfanilamide (I to V).

Bock and Frenzel[495] ligated the splenic vein in 5 rabbits and produced inhibition of the bone marrow. The erythrocytes returned to normal in ten weeks, but the leukocytes did not do so until thirty-five weeks had passed and only after treatment had been given. Hölderlin[496] injected into rabbits a 5 per cent sterile solution of milk casein, Bacillus coli vaccine, horse serum and histamine at intervals of three to four days. When the animals were sensitized, an injection was given and the bone marrow examined three to five days later. Some specimens showed "irritation" and others exhaustion. Serum was most toxic and milk casein and histamine the least. There appeared to be no parallelism between the condition in the bone marrow and the picture of the peripheral leukocytes.

Rhoads and Miller[497] studied 69 patients with idiopathic progressive anemia and found that the sternal bone marrow suggested a pentavalent classification. The first group showed replacement of hemopoietic tissue by a cellular structure composed of megakaryocytes in various stages of development. In the second group the marrow was sclerotic. The remaining three groups showed various degrees of cellularity of the marrow, aplastic, active and hyperplastic. In the latter groups there was failure of the hemopoietic cells to mature beyond an early undifferentiated stage. A relation to agranulocytosis was postulated.

After roentgen therapy, Meller, Gottlieb and Brauner[498] found, by means of repeated sternal punctures, that there is an excitation of the young elements of the bone marrow during the first few hours, with subsequent maturation. There is a considerable increase of adult granulocytic elements (segmented polymorphonuclear neutrophils). The increase in 1 case was from 284 per thousand to 628 per thousand

494. The roman numerals after the names of the drugs refer to the diseases that have just been listed.

495. Bock, H. E., and Frenzel, B.: Splenogene Knochenmarkhemmung: Tierexperimentellen Beweis, Klin. Wchnschr. **17**:1315, 1938.

496. Hölderlin, H.: Knochenmark und Blutbild bein sensibilisierten Tier, Virchows Arch. f. path. Anat. **302**:118, 1938.

497. Rhoads, C. P., and Miller, D. K.: Histology of the Bone Marrow in Aplastic Anemia, Arch. Path. **26**:648 (Sept.) 1938.

498. Meller, O.; Gottlieb, F., and Brauner, R.: The Importance of the Indications from Sternal Puncture in Roentgen Therapy, Radiology **31**:149, 1938.

twenty-four hours after irradiation. The authors feel that the myelogram is a valuable guide during the course of roentgen therapy.

In 13 cases of multiple myeloma, Rosenthal and Vogel [499] found no characteristic blood picture. Thrombocytopenia was present in 5 cases. In myelocytic myeloma, myelocytes and myeloblasts appeared in the blood stream. While roentgen observations, hyperproteinemia, the "formol gel" reaction and Bence Jones proteinuria were of diagnostic aid in some cases, the finding of myeloma cells on sternal puncture revealed the underlying condition.

HEMATOLOGIC METHODS

The time of erythrocyte regeneration after hemorrhage from peptic ulcer is constant, according to Schiødt,[500] being thirty-three days. The rate of increase varies with the level of the red blood cell count after hemorrhage ceases, but the rate of production is always that normally required for balance. The rate of destruction is decreased in proportion to the amount of blood lost, since old as well as young cells are removed by bleeding and the new cells entering the circulation are not ripe for destruction.

Ettori and Grangaud [501] have modified the Ponder and Saslow method of estimating the total cell volume, which depends on determination of the concentration of a known quantity of free hemoglobin added to a known volume of whole blood. Their values for normal subjects varied between 40 and 50 per cent, with an error said to have been within 1 per cent.

Studies of the blood volume have been reported by Gibson and his associates,[502] employing the azo dye Evans blue, the spectrophotometer and the Evelyn photoelectric microcolorimeter. The method was said to give constant values for plasma volume within a range of plus or minus 2.5 per cent. For clinical research the microcolorimeter was preferred by the authors to the spectrophotometer, because of its relative simplicity, speed, economy and absence of subjective error. An analysis of colorimetric methods of obtaining plasma volume determinations was

---

499. Rosenthal, N., and Vogel, P.: Value of the Sternal Puncture in the Diagnosis of Multiple Myeloma, J. Mt. Sinai Hosp. **4**:1001, 1938.

500. Schiødt, E.: A Standard for Estimating Blood Regeneration After Hemorrhage, Acta med. Scandinav., 1938, supp. 89, p. 157.

501. Ettori, J., and Grangaud, R.: Sur la détermination du volume globulaire: influence des anticoagulants sur le volume globulaire mesuré par photométric, Compt. rend. Soc. de biol. **128**:181, 1938.

502. Gibson, J. G., Jr., and Evans, W. A., Jr.: Clinical Studies of the Blood Volume: I. Clinical Application of a Method Employing the Azo Dye "Evans Blue" and the Spectrophotometer, J. Clin. Investigation **16**:301, 1937. Gibson, J. G., Jr., and Evelyn, K. A.: Clinical Studies of the Blood Volume: Adaptation of the Method to the Photoelectric Microcolorimeter, ibid. **17**:153, 1938. Thomson, Hirsheimer, Gibson and Evans.[164]

published by Gregersen.[503] He criticized, on grounds of inaccuracy, the ordinary dye and colorimetric methods and advocated the use of the blue dye T-1824, with spectrophotometric determinations.

A calibrated micropipet with a 50 cubic millimeter capacity has been devised by Kato.[504] It is adapted for small quantities of blood and is applicable to a number of hematologic procedures, including determinations of the sedimentation rate and the volume of packed cells and microprecipitation tests for syphilis.

A number of papers dealing with the factors influencing sedimentation of erythrocytes and questioning methods of so-called correction of the rate for changes in cell volume have been published during the past year. Tiffeneau and Gysin [505] separated plasma, by ultrafiltration, into two fractions. The rate of settling of the red blood cells was decreased in the nonprotein fraction, whereas it was increased in the protein fraction. In the case of specimens of blood possessing different sedimentation rates, the rates of settling become the same when the nonprotein plasma fractions are used but vary when the protein fractions are employed in accordance with the rates of the specimens from which the fractions were obtained. On the other hand, the use of cells from different specimens of blood does not influence the rate. The authors also concluded that plasma protein accelerates the sedimentation rate in accordance with its molecular size. Schuster [506] reported his observations in 121 cases and found that polycythemia retards the sedimentation rate, whereas anemia accelerates it but that no charted system of correction for changes in the blood count is satisfactory. He advocated adjustment of the count to a standard concentration by removal or addition of plasma. Bouton,[507] however, stated, from his observations on patients after malaria inoculation that the relative number of erythrocytes in the plasma has a negligible effect per se on the rate of settling of the red blood cells and that such an effect as may exist is inconstant. He concluded that attempts at compensation for anemia represent pseudoaccuracy and that the unmodified sedimentation curve, graphi-

503. Gregersen, M. I.: An Analysis of Colorimetric Methods in Relation to Plasma Volume Determinations, J. Lab. & Clin. Med. **23**:423, 1938.

504. Kato, K.: Combination Microhemopipette for Determination of the Sedimentation Rate, Packed Cell Volume and Fragility of Erythrocytes, Especially Adapted for Use in Children and Small Laboratory Animals, J. Lab. & Clin. Med. **23**:980, 1938.

505. Tiffeneau, R., and Gysin, O.: Rôle des protides plasmatiques dans la vitesse de sédimentation des hématies: Influence du volume des molécules protidiques, Compt. rend. Soc. de biol. **126**:1160, 1937.

506. Schuster, N. H.: Sedimentation Rate in Relation to the Red Cell Count: The Problem of Correction, Tubercle **19**:529, 1938.

507. Bouton, S. M., Jr.: Erythrocyte Sedimentation and Anemia: A Preliminary Report, J. Lab. & Clin. Med. **23**:519, 1938.

cally recorded, is a valuable diagnostic aid. Hambleton and Christianson [508] studied the sedimentation rate of blood adjusted with isotonic solution of sodium citrate to cell volumes of 25, 29, 35, 40 and 45 per cent. Their results indicated to them that correction for the cell volume is, in general, of advantage for blood with a volume of packed cells in excess of 45 per cent but that correction was undesirable when the volume was subnormal. Cutler, Park and Herr [509] stated that anemia has little or nothing to do with the aggregation or sedimenting phase of the sedimentation phenomenon but that it is concerned with the terminal or packing phase. Consequently, according to these authors, correction for anemia by charts or by adjustment of plasma to obtain a normal cell volume is inaccurate, and interpretation should be based on the shape of the sedimentation curve, as well as on the extent of the fall at one hour. Hynes and Whitby,[510] on the other hand, have supported the principle of the Wintrobe and Landsberg method of correction of the sedimentation rate for anemia by means of a logarithmic chart but have put forward a new correction chart based on a wider range of rates than that employed by Wintrobe and Landsberg. They advocated expressing the sedimentation rate in general terms rather than as an exact figure. Observations of the sedimentation rate for children with nutritional anemia led Smith [511] to conclude that the blood in these cases does not behave as does diluted normal blood. A further report was published by Lee [512] on an automatic photographic recorder of erythrocyte sedimentation.

---

508. Hambleton, A., and Christianson, R. A.: A Simplified Method of Correcting the Sedimentation Rate for the Effect of Cell Volume, J. Lab. & Clin. Med. **23**:860, 1938.

509. Cutler, J. W.; Park, F. R., and Herr, B. S.: The Influence of Anemia on Blood Sedimentation, Am. J. M. Sc. **195**:734, 1938.

510. Hynes, M., and Whitby, L. E. H.: Correction of the Sedimentation Rate for Anaemia, Lancet **2**:249, 1938.

511. Smith, C. H.: Sedimentation Rate in Nutritional Anemia of Infants and Children: Its Response to Treatment with Iron (Ferrous Sulfate), Am. J. Dis. Child. **56**:510 (Sept.) 1938.

512. Lee, T.: The Sedimentometer: A Photographic Recorder of the Suspension Stability of the Erythrocyte, Am. J. M. Sc. **195**:729, 1938.

News and Comment

Biological Photographic Association.—The ninth annual convention of the Biological Photographic Association will be held September 14 to 16 at the Mellon Institute for Industrial Research, Pittsburgh. The program will be of interest to scientific photographers, scientists who use photography as an aid in their work, teachers in the biologic fields, technical experts and serious amateurs. It will include discussions of motion picture and still photography, photomicrography, color and monochrome films and processing, all in the field of scientific illustrating. Up-to-date equipment will be shown in the technical exhibit, and the print salon will display the work of many of the leading biologic photographers in the United States and abroad.

The *Biological Photographic Association Journal* is published quarterly and constitutes a volume of about 250 pages, which is furnished free to members. Membership privileges include an authoritative question and answer service and the right to borrow loan albums and exhibits of scientific prints for study and display.

Further information about the association and the convention may be obtained by writing the secretary of the Biological Photographic Association, University Office, Elizabeth Steel Magee Hospital, Pittsburgh.

Book Reviews

Kvantitative urinsediments bestemmelser: Et bidrag til belysning af proteinspørgsmaalet ved nefritisdiäten. By Asger Naeraa. Pp. 191. Copenhagen: Nyt Nordisk Forlag-Arnold Busck, 1936.

The author gives in this book, or thesis, for which he was awarded the degree of Doctor of Medicine at the University of Copenhagen, an illustration of the effect of the protein diet in the treatment of nephritis. He calls the work "Quantitative Determinations of Urinary Sediments."

In the introduction he mentions the three generally accepted types of Bright's disease, their characteristic symptoms being (1) hematuria, (2) albuminuria and (3) hypertension. But it is on the classification made by Addis (hemorrhagic, degenerative and arteriosclerotic), with some modifications of his own, that he bases his clinical experiments. With sediment counts he finds it possible to follow the course of inflammatory renal conditions from day to day.

He soon found much uncertainty concerning the following phases of the problem of sediment counts: (1) the fluctuation of the sediment contents under different normal physiologic conditions, (2) the quantity of sediment in persons with benign albuminuria and (3) the influence of the usual hospital medicaments, especially salicyl preparations.

The first part of the book is devoted to the examination of these problems, the discussion of the organic components of the sediment and the description of the technic used by the author.

The second part deals with the main issue, the significance of the diet during the treatment of acute hemorrhagic nephritis, with special reference to the possible injuriousness of protein. Naeraa points out that the uncertainty in regard to the influence of the diet has caused most patients with acute nephritis to be kept on a general diet for a feverish condition and that a remarkable number of physicians still adhere to the slogan of Crestien *"Le lait ou la morte."* The result is that many patients suffer from undernourishment.

After examination of many discussions of the problem, the author finds that the main reason for disagreement is that the writers do not keep its different phases separate. He therefore classifies the main questions as follows: (1) the possible injuriousness and irritating effect of protein during acute inflammation of the kidney; (2) the possible injurious effect of protein during chronic nephritis, especially arteriosclerotic nephritis, and (3) the possible injurious influence of protein when renal insufficiency is present, as shown by the nonprotein nitrogen of the blood. The only one of these questions on which all agree is that protein must not be given during acute renal insufficiency.

After a short review of the importance of the different symptoms, followed by an outline and diagrams of the course of hemorrhagic nephritis, the author describes his experiments with a number of patients. Only males were selected, in order that urine free from other elements might be obtained. He first gave 9 patients a diet of 2,750 calories, with a protein content of 40 Gm. After the hematuria became constant a change was made to 3,000 calories, with 125 Gm. of protein. The two diets were thereafter given alternately, the period of each lasting from eight to nineteen days. One patient was given the high protein diet only, and 3 were kept on the usual low protein nephritic diet. Daily counts of the urinary sediments were kept in all cases. The description of each of the experiments is illustrated by graphs and tables, and an abstract of each case is printed at the end of the book.

In conclusion, Naeraa gives the following summary of his studies and experiments:

1. The Addis method of counting the number of cells and casts in the urine, as well as the author's modification of it, gives a true estimate of these elements.

2. When the urine has been separated and Quensel's color fluid added, the formed elements (if the urine is concentrated) will keep for several days.

3. Sediment determinations for 70 apparently well persons aged from 20 to 40 gave the following values for twelve hour night urine:

Erythrocytes	Leukocytes and Epithelial Cells	Casts
0-1,100,000	48,000-4,000,000	0-8,700
130,000	1,000,000	1,900

All casts were hyaline.

In the case of 10 persons aged from 55 to 90 the sediment deviated from normal. Casts were found in great number, up to 15,000.

4. Examination of the day urine from 21 of the 70 persons mentioned did not show any difference in the amount of formed elements from that of the night urine. The nephritic patients gave indication of greater bleeding during the day than at night.

5. Examination of the sediments from normal persons exposed to great exertion gave no indication of any exertion hematuria. On the other hand, the number of casts may rise as high as that found in patients with nephritis. All casts are hyaline, even though the precipitation of crystals may give them a deceptive resemblance to granular casts. The number of casts seems to increase with the duration of the exertion.

6. Salicyl preparations, even in ordinary therapeutic doses, will cause an increase in the number of casts. After only 1 Gm. of magnyl has been given there may be as many as 50,000 casts in twelve hour night urine. After a salicylate has been taken daily for a longer time, this number may increase to several hundred thousand. The casts are all hyaline. It is presumed that the casts increase in number because the salicyl radical disengages the protein substance normally contained in the urine. There is no indication that salicyl preparations damage the kidneys.

7. Examination of the night and day urine of 11 boys with benign albuminuria showed that the sediment did not contain an abnormal number of erythrocytes or leukocytes, but that off and on the number of hyaline casts was increased.

8. The highest recorded number of erythrocytes found in the night urine during the acute stage of hemorrhagic nephritis is 10,000,000,000, which means about 2 cc. of blood.

During the acute stage of hemorrhagic nephritis the patients have real pyuria. Sometimes they secrete as many white as red blood corpuscles.

10. A graph for sediment counts carried out daily during the course of hemorrhagic nephritis shows great fluctuation in the number of erythrocytes, even when they are decreasing. The white corpuscles, on the other hand, show less fluctuation. Great increase of erythrocytes is often seen, but does not mean any aggravation of the process. An increase of the white blood corpuscles accompanies aggravation. The activity of the renal inflammation ought therefore always to be estimated from the number of leukocytes and epithelial cells in the urine, and not from the degree of hematuria.

11. The number of casts during the course of hemorrhagic nephritis varies; yet this is not a measuring stick of renal inflammation. The number of hyaline casts, on the other hand, is always in a certain proportion to the degree of albuminuria.

12. Hyaline casts may be formed as soon as the urine is sufficiently concentrated. The proteins which appear in the urine under normal circumstances will

be disengaged at the isoelectric point. This disengagement takes place where the urine is concentrated by retrograde resorption, namely, in the tubules where the casts are formed.

13. General clinical impressions, laboratory examinations and daily sediment counts made on patients with hemorrhagic nephritis who have received diets alternately poor and rich in proteins have not indicated that a diet rich in meat has damaged the kidney.

The general impression during the experiments was that the renal disease seemed to improve with food rich in proteins.

This monograph contains a large store of information concerning the significance of sediment counts of the urine and the influence of various factors on them. The careful study of the course of patients with nephritis requires some method of measuring the inflammation in the kidney. A study of the problem as outlined by Naeraa is desirable and profitable.

The Clinical and Experimental Use of Sulfanilamide, Sulfapyridine and Allied Compounds. By Perrin H. Long, M.D., Associate Professor of Medicine, The Johns Hopkins University, and Eleanor A. Bliss, Sc.D., Fellow in Medicine, The School of Medicine, The Johns Hopkins University. Price, $3.50. Pp. VII + 319, with 6 charts. New York: The Macmillan Company, 1939.

The entire medical profession is indebted to the authors of this monograph. For in it Drs. Long and Bliss have presented a clear and interesting statement of current opinion regarding the clinical usefulness of sulfanilamide and allied compounds. Their own knowledge of the subject is, of course, authoritative.

The book represents an amazing bit of work, for it must have been written during a time when the authors were under great pressure, when new knowledge was coming to light almost every day and when current literature, which had to be read and sifted critically, was growing with prodigious speed. Evidently the authors have felt a strong sense of responsibility for making the information which they have released as recent as possible. The first printing appeared in June, and yet a special addendum was inserted so late as April 25, and the preface was written on February 14. The book, therefore, is as up-to-date as they could make it.

It begins with a delightfully written chapter giving the history of sulfanilamide therapy. Then follow seven chapters which deal with the chemotherapy of experimental bacterial infections, the comparative pharmacology of sulfanilamide and allied compounds, the mode of action of these drugs, their clinical use and, finally, their clinical toxic manifestations. At the end of each chapter is a bibliography; toward the end of the book, an index of authors, and at last, an unusually complete subject index.

The part of the volume which deals with clinical treatment will appeal most to practicing physicians. The authors show admirable restraint and take care to be conservative rather than too enthusiastic. They mention the variety of diseases which are being treated with sulfanilamide, the customary dose of the drug in common use and the therapeutic results which seemingly have been obtained in their own experience or in that of others.

They discuss debatable matters in connection with sulfanilamide therapy frankly and with open mind; they point out gaps in present knowledge; they look to the future. Not only do they make any reader sense the importance of the discovery of sulfanilamide and explain how he may use the drug or its allied compounds with reasonable intelligence, but they stimulate imagination by suggesting the diverse ways by which, in the future, chemotherapy is likely to become increasingly significant. On the whole, the book is one of the most important books of the year. Every physician should read it.